Journal of
DAIRY SCIENCE

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Purdue University, Lafayette, Indiana

August 4, 1958

JOURNAL OF DAIRY SCIENCE

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Advertising

Advertising plates, copy, insertion orders, contracts, and requests for information relating to advertising in the Journal should be addressed to The Garrard Press, 510-522 North Hickory Street, Champaign, Illinois.

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Printed by The Garrard Press, 510-522 North Hickory Street, Champaign, Illinois. Entered as second-class matter at the post office at Champaign, Illinois, under the act of March 3, 1879.

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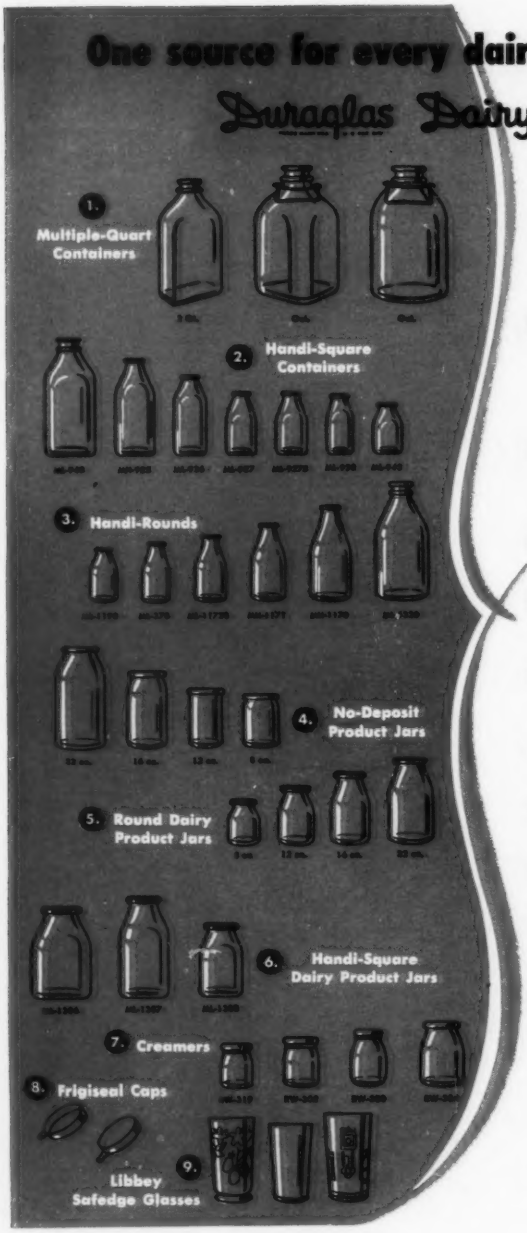

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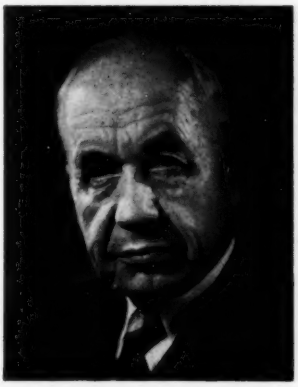
PEOPLE AND EVENTS

C. E. Lee, A.D.S.A. Charter Member, Dies in Milwaukee



C. E. Lee

C. E. LEE, 82, a charter member of the American Dairy Science Association, died March 16, in Milwaukee.

Born in Albion Prairie, Wis., Mr. Lee was graduated from North Dakota State Agricultural College in 1891 and received his M.S. degree from the Univ. of Wisconsin in 1909. He served on the Univ. of Wisconsin staff until 1915, when he became assistant Dairy and Food Commissioner and Dairy Specialist for the State of Wisconsin until 1920.

In 1920, he joined the Gridley Dairy Co. (now a division of Borden's), Milwaukee, as manager of the laboratory and manufacturing operations, a position he held until his retirement in 1940. From 1940 until 1953 he did consulting work.

Mr. Lee was present at the first meeting of a group of dairy instructors and investigators who discussed the idea of forming an association. There are now four living Charter Members of the Association.

Mr. Lee is survived by his widow, Elizabeth; two sons, H. J. LEE, Milwaukee, Wis., and H. C. LEE, Richmond, Va., and a daughter, Mrs. J. P. RUSSELL, New Hartford, N. Y.

Forage Evaluation Symposium to be Held at Purdue Aug. 4

A Forage Evaluation Symposium will be held Aug. 4 in conjunction with the annual meeting of the American Society of Agronomy at Purdue Univ., Lafayette, Ind. Other organizations participating are the American Society of Animal Production, the American Society of Range Management, and the A.D.S.A.

The following papers will be given:

1. The Significance of and Techniques Used to Measure Forage Intake and Digestibility—M. E. MCCULLOUGH, Georgia Agricultural Experiment Station, Experiment.
2. The Use of Esophageal Fistula, Lignin, and Chromogen Techniques for Studying Selective Grazing and Digestibility of Range and Pasture by Sheep and Cattle—W. C. WEIR, J. H. MEYER, AND G. B. LOFGREEN, Univ. of California, Davis.
3. Intake and Digestibility Techniques and Supplemental Feeding in Range Forage Evaluation—L. E. HARRIS AND C. W. COOK, Utah State Agricultural College, Logan.
4. Animal Variation and Measurements of Forage Quality—G. O. MOTT, Purdue Univ., Lafayette, Ind.
5. Progress in Feed Evaluation—MAX KLEIBER, Univ. of California, Davis.
6. Relationship of Digestion End-Products to the Energy Economy of Animals—J. C. SHAW, Univ. of Maryland, College Park.
7. Yield and Quality of Forage Under Grazing and Clipping—R. E. BLASER, Virginia Polytechnic Institute, Blacksburg.
8. What Is Forage Quality from the Animal Standpoint?—J. T. REID, Cornell Univ., Ithaca, N. Y.

B. F. Castle Honored by MIF

COL. B. F. CASTLE, retired executive vice-president of the Milk Industry Foundation, was honored recently at a testimonial dinner tendered by the MIF board of directors.

A. G. MARCUS, president and general manager of Dairymen's Association, Ltd., Hawaii, and past president of MIF, paid tribute to Col. Castle for his services to the Foundation during the last 18 years. In particular, Mr. Marcus credited Castle with saving the retail home-delivery milk business from being regulated out of existence during World War II.

F. B. BALDWIN, vice president of Abbotts Dairies, Inc., and chairman of the MIF advisory committee, served as master of ceremonies for the program. Other past presidents of the Foundation who paid tribute to Col. Castle included C. R. BROCK, Brock-Hall Dairy Co., Hamden, Conn.; T. K. HAMILTON, Diamond Milk Products, Inc., Columbus, O., and F. H. KULLMAN, JR., Bowman Dairy Co., Chicago. The present MIF president, HUBERT GARRECHT of Klinke-Reed Dairies, Inc., Memphis, Tenn., also spoke.

Col. Castle, a West Point graduate, was one of the first U.S. Army fliers to see combat action in World War I. After that, he became Eastern European representative and later assistant to a vice-president of the Irving Trust Co., N. Y.

EXCITING DAYS

at Foremost Dairies

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And an instant whole milk is now being produced. This new product affords all the convenience of instant nonfat milk—and all the butterfat of fresh whole milk.

Another new product at Foremost is its "Fresher Tasting" evaporated milk—the first really new evaporated milk in over half a century.

In addition to these efforts to expand the market for dairy products, Foremost has launched a vigorous educational program to demonstrate new uses, new times, new places for American families to enjoy existing dairy products.

So you can see, there's a lot to be excited about these days at

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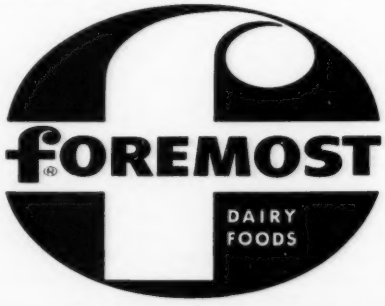

From 1926 to 1940, he was successively president of the First Federal Foreign Banking Corporation; president of the Great Lakes Aircraft Corp., and vice-president and director of the Administrative and Research Corp.

When MIF was merged with the International Association of Milk Dealers in 1946, Col. Castle was named executive director of the combined organization. In 1953, he was appointed executive vice-president in charge of federal affairs.

The Congressional Medal of Honor was awarded posthumously to Col. Castle's only son, BRIGADIER GEN. F. W. CASTLE, U. S. Air Force, for leading mass air attacks against Germany during World War II. Castle Air Force Base in California is named for him.

E. Jensen Promoted by Borden's

EDWARD JENSEN has been named assistant supervisor of country plants for the Borden Company's Farm Products Division in New York State.

T. W. TALLEUR succeeds Mr. Jensen as district superintendent of District One, which embraces country plants in Cochecton and Warwick, N. Y., and Sussex County, N. Y.

Mr. Talleur has been with Borden's since 1928 and Mr. Jensen has been with the company since 1933.

USDA Uses Automation to Study Cow Metabolism

Automation, combined with ingenious equipment and instruments, is making possible long-needed fundamental research on cow metabolism.

A new highly-automated, energy-metabolism laboratory, located at the USDA's Agricultural Research Center, Beltsville, Md., was recently put into operation to study such problems as how much of a cow's feed goes into milk.

This laboratory is large enough for the study of six cows at a time in individual airtight chambers under controlled conditions. It is the biggest and most automatic facility of its kind in the world.

The researchers hope to learn how much heat a cow produces, and how much food she digests and metabolizes to maintain her body. The remainder will represent the food that produces milk and body gain.

The studies also are aimed at devising better analytical methods, so that more accurate evaluation of feeds can be made. Samples of all feed are analyzed, to determine their crude fiber, fat, protein, nitrogen-free extract, and carbon. In addition, other methods of analysis are being developed to improve correlation between chemical analyses and feeding value.

Various forages, alone or in combination with concentrates, will be fed to discover the relative nutritional values of individual feeds and complete rations.

Pennsylvania News

E. C. FOLKERS, general manager and vice-president of Meadow Gold Dairies, Inc., Pittsburgh, has been elected president of the Pennsylvania Association of Milk Dealers.

A graduate of the Univ. of Illinois with a M.S. degree from Pennsylvania State Univ., Folkers served as vice-president and assistant general manager of the Meadow Gold Dairies from 1942 to 1952. He has been general manager of the company since 1952.

West Virginia News

The 83rd Annual Convention of the Ayrshire Breeders Association was held in May at Jackson's Mill, Weston, W. Va.

High lights of the event were an Open House at the West Virginia Univ. Dairy Farm, home of the Reymann Memorial Ayrshires; a tour of the University's new Medical Center; a tour of the Clarksburg Carnation Company evaporating plant; a dance and other entertainment, and a panel discussion.

The panel discussion, on the topic of "The Value of Breed Improvement Programs in Merchandising," was moderated by I. D. PORTERFIELD, head of the West Virginia Univ. Dairy Dept. Panel members were RICHARD SEARS, G. A. BOWLING, and JACK NISBET.

West Virginia's GOV. CECIL UNDERWOOD was the featured speaker at the convention banquet. At the banquet, MISS DORIS CHADBURN was awarded the Distinguished Service Citation for some 30 years of service as manager of the Association's central office.

A dairy cattle Judging School was held at West Virginia Univ. in April under the direction of R. D. STEWARD, secretary of the Guernsey Breeders Association, and I. D. PORTERFIELD, head of the Dairy Dept. at W.V.U.

The event was held at the request of the state Purebred Dairy Cattle Club, to train county agents, vocational agriculture teachers, and breeders in the fundamentals of dairy cattle judging. The course was designed to provide more competent judges for community and county fairs held throughout the state and to provide a refresher course for experienced judges.

Ohio State News

The Dept. of Dairy Technology has released its third volume of *Research in Review*, a book covering publications of department staff members for the years 1956 and 1957.

HENRY DE HAMEL, industrial engineer of the Kroger Co., Cincinnati, spoke on "Work Simplification in the Dairy Plant" at the May meet-

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ings of the four Ohio Dairy Technology Societies—Maumee Valley, Northeastern Ohio, Central Ohio, and Cincinnati.

PHILIP HINTZ, instructor in dairy technology, has accepted a position with the New Product Research Laboratory of the Borden Company, Syracuse, N. Y.

Events and judges have been chosen for the dairy products section of the Ohio State Fair to be held Aug. 22-29.

Events in the manufacture and judging of dairy products will be Swiss cheese making for block and wheel type cheese, cottage cheese making, ice cream judging, milk judging for college-trained persons, and milk judging for high school students and industry persons without formal training in judging.

Official judges will be R. E. HARDELL, Monroe, Wis., for Swiss cheese, and J. M. JENSEN, Michigan State Univ., East Lansing, for cottage cheese.

The governing committee of the Ohio Dairy Products Research Fund met in May at the Dept. of Dairy Technology. This committee reviews and advises on the department's entire research program and oversees the operation of the Ohio Dairy Products Research Fund and the Everett Antrim Fund.

Members of the committee include O. C. WOOLPERT, director of the Ohio State Univ. Research Foundation; K. S. CAMPBELL, field director of the Ohio State Development Fund; E. WALTERS, dean of the O.S.U. Graduate School; L. L. RUMMELL, dean of the O.S.U. College of Agriculture; J. H. ERB, The Borden Co., Columbus; ALLEN FRENCH, French-Bauer Co., Cincinnati; H. F. MEYER, Meyer Dairy Products Co., Cleveland, and F. S. GREINER, executive secretary of the Ohio Dairy Products Association.

The Fifth Annual Clinic for managers and accountants was conducted in May by the Ohio Dairy Products Association. Speakers included C. W. DOLL, Model Farms Dairy, Louisville, Ky.; PAUL BECK, International Association of Ice Cream Manufacturers, Washington, D. C.; J. M. FORBIS, E. B. McClain Co., Memphis, Tenn., and T. K. HAMILTON, Diamond Milk Co., Columbus.

F. B. Morrison Dies April 7

Word has been received of the death April 7 of DR. F. B. MORRISON, Sarasota, Fla., retired head of the Dept. of Animal Husbandry, Cornell Univ., Ithaca, N. Y.

Born May 19, 1887, at Fort Atkinson, Wis., Dr. Morrison received his B.S. degree from the Univ. of Wisconsin in 1911, and D.Sc. degrees from the Univ. of Vermont and the Univ. of Wisconsin in 1947 and 1950, respectively.

Dr. Morrison became Director of the Cornell Univ. Agricultural Experiment Station in 1927 after some 16 years on the faculty at the Univ. of Wisconsin. His last job at Wisconsin was that of assistant director of the Experiment Station.

In 1928, he became professor of animal husbandry and animal nutrition, and head of the Animal Husbandry Dept., at Cornell, a position he held until 1945. From 1945 to 1955 he gave up his administrative responsibilities, but continued his teaching and research.

During his long career, Dr. Morrison participated in different U. S. overseas projects in Germany, the Philippines, Argentina, and Venezuela.

He was the author of two well-known books, *Feeds and Feeding* and *Feeds and Feeding, Abridged.*

In 1946, Dr. Morrison established an annual award which enables the American Society of Animal Production to give a substantial cash prize each year to a scientist selected by the Society for his outstanding research in livestock production. In 195, Dr. and Mrs. Morrison endowed a fellowship at Cornell Univ. to foster the training of graduate students, especially those from foreign lands, in the field of livestock feeding.

He is a past president of the American Society of Animal Production and a member of the American Dairy Science Association, Alpha Zeta, Sigma Xi, Phi Lambda Upsilon, and Phi Kappa Phi.

Survivors include his widow, MRS. ELSIE BULLARD MORRISON, and two sons, ROGER B. and SPENCER H. MORRISON.

LaMaster Takes Post in Brazil

J. P. LAMASTER, professor emeritus of dairying at Clemson Agricultural College, Clemson, S. C., has accepted a position as livestock advisor to the U. S. Mutual Security Operations Mission in Rio de Janeiro, Brazil. This program is conducted under the auspices of the U. S. International Cooperation Administration.

Utah Dairy Tech. Group Meets

The Utah Dairy Technology Society met at Salt Lake City in May with L. A. BLACK, chief milk sanitarian for the U. S. Public Health Service, as principal speaker.

At the meeting the following officers were elected: S. A. ROSE, Paramount Ice Cream Co., Ogden, president; WILLIAM GODFREY, Hi-Land Dairy Association, Murray, vice-president, and P. B. LARSEN, Dairy Industry Dept., Utah State Univ., Logan, secretary-treasurer. Directors elected are ROBERT DOBSON, Wever Central

A report from MoorMan's

Here is the latest photo of MoorMan's Dairy Research Unit. Soon the efficient operation of the unit—started 4 years ago—will be increased with the erection of a new maternity and utility barn.

This is one of the world's largest privately owned dairy farms dedicated solely to research work. There are facilities for controlled experiments with identical twins as well as for the conventional group allotment tests.

Nutrition studies are carried out on animals of all ages and cover a broad range. These studies include work on such things as trace minerals, hormones, rumen organisms, antibiotics, vitamins, drugs and certain palatability factors.

In addition, work here and in field experiments is being conducted to determine the effect various management practices associated with feeding have on the growing heifer. These tests include levels and systems of feeding and their effect on growth, reproduction, general health, and finally on level and persistency of production.

If you are going to be in the vicinity of Quincy we'd like very much to show you the unit and answer any questions you may have about it and its operation. If you have the time we'd like, also, to have you visit our Research Laboratories and spend some time in our modern factory. If you can visit us, drop us a line a few days in advance so that we can arrange to have one of our men show you around. If you're with us around noon we'd like to have you as our guest for lunch in our new cafeteria.

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Quincy, Illinois

Dairy Association, Ogden; ALVIN FERRE, Ogden City Health Dept., and ANCIL WINGER, Beatrice Foods Co., Orem.

Arizona Receives A.D.A. Grant

A $4,500 grant has been awarded to the Univ. of Arizona by the American Dairy Association. The award is for the first year of a three-year study of the relationship between the composition of milk beverages and consumer preference.

The study will be under the direction of J. W. STULL, Dept. of Dairy Science, and J. S. HILLMAN, Dept. of Agricultural Economics.

Completed Theses

M.S. Degree

D. R. ERICKSON—Efficiency of certain Flavone-type compounds as antioxidants for dairy products. Oregon State College, Corvallis.

E. A. ZOTTOLA—Effect of certain manufacturing methods on the physical characteristics of butter. Oregon State College, Corvallis.

Ph.D. Degree

W. E. SANDINE—The physicology and toxonomy of the carbon dioxide–producing lactic acid streptococci. Oregon State College, Corvallis.

Bush Joins Oklahoma Staff

LYNN BUSH has joined the faculty of the Oklahoma State Univ. Dairy Dept. as associate professor. He replaces M. RONNING, who resigned to take a position at the Univ. of California.

Dr. Bush received his B.S. degree from the Univ. of Kentucky, his M.S. degree from the Ohio State Univ., and his Ph.D. degree from Iowa State College. During the past three years, he has been a research associate with the Iowa Agricultural Experiment Station.

At Oklahoma, he will be in charge of dairy cattle nutritional research for the Oklahoma Experiment Station.

N.D.C. News

The National Dairy Council has announced the opening of two new affiliated Dairy Council units. They are the Dairy Council of Chattahoochee Valley, Columbus, Ga., and the Dairy Council of Macon–Warner Robins, Macon, Ga.

Officials of the Chattahoochee Valley unit include L. N. SOLOMON, JR., president; GEORGE HATCHER, vice-president; W. J. CAUSEY, secretary-treasurer, and MRS. M. G. NEWBERRY, executive director.

For the Macon unit, officials include A. F. RANDALL, president; J. C. WEBB, vice-president; J. R. KINNETT, secretary-treasurer, and MRS. JOHN LANFORD, executive director.

Speakers for the 1958 N.D.C. Summer Conference, to be held in Chicago, June 30—July 2, have been announced.

CLARK TIBBITTS, assistant director of the staff on aging, U. S. Dept. of Health, Education, and Welfare, will speak Monday morning on the subject, "What Is the Senior Citizen Problem?"

Monday afternoon, three speakers will be featured in a session on "Basing Opinions on Facts." The speakers are R. E. SHANK, Washington Univ.; R. E. OLSON, head of the Dept. of Biochemistry and Nutrition, Univ. of Pittsburgh, and ZOE E. ANDERSON, N.D.C. director of nutrition research.

Other speakers to participate in the conference include G. V. BOHMAN, chairman of the Dept. of Speech, Wayne Univ., Detroit, Mich.; WARREN GUTHRIE, chairman of the Dept. of Speech, Western Reserve Univ., Cleveland, O.; R. J. SPEIRS, president of Abbotts Dairies, Inc., Philadelphia; SHELBY SCHILD, Div. of Information Programs, USDA, and H. E. CLARKSON, consultant to the American Institute of Banking, Chicago.

Cherry-Burrell to Move Headquarters

The general offices of Cherry-Burrell Corporation will be moved from Chicago to Cedar Rapids, Ia., about July 1, according to a recent announcement by H. H. CHERRY, JR., president.

The general offices will be located in the two-story administration building at the company's new, nine-acre factory at 2400 Sixth street, S.W., in Cedar Rapids.

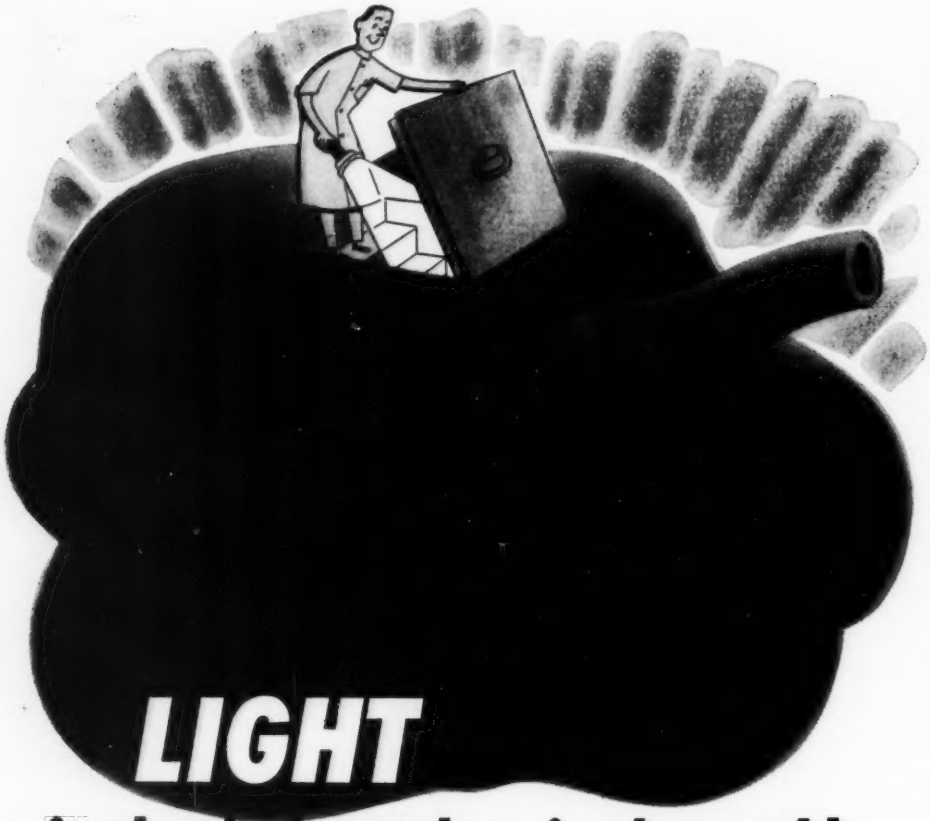
The corporation's other large manufacturing plant at Little Falls, N. Y., was enlarged in 1955 and is now being remodeled to accommodate the administrative and engineering activities connected with the Little Falls operation.

Remaining in its present location at 822 West Washington blvd., Chicago, will be the Chicago Sales Branch. This unit will be augmented to provide services previously available to customers from the nearby general offices.

Montana News

The Dairy Industry Dept. of Montana State College, Bozeman, will hold its Dairy Production Day on Nov. 19, 1958. Central theme of the event will be a statewide forage show and forage program.

Forage samples will be judged by competent judges and prizes will be awarded for the best samples exhibited. All samples will be analyzed chemically and each exhibitor will receive the results of the chemical analysis.



in the darkest place in the world...

The inside of a cow's stomach was "the darkest place in the world" until scientists opened trapdoors to let in the bright light of research. They are learning the secrets of the rumen and its complicated microbiological processes.

These studies are revealing new facts about the function of urea in mixed feeds for dairy cattle and other ruminants. The micro-organisms in the rumen use urea to manufacture protein. Urea also helps them break down cellulose and other fiber and get more feed value out of roughage. Thus urea helps ruminants produce more meat and milk at low cost.

More and more leading feed manufacturers are using PROCADIAN® Urea as a source of protein in high-quality complete feeds and supplements. As a result of research, PROCADIAN Urea is helping our cud-chewers to become better producers of protein and profits!

NITROGEN DIVISION
40 Rector Street,
New York 6, N.Y.

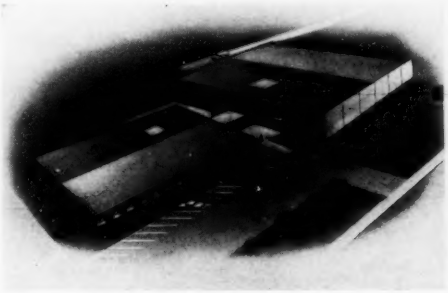
Allied Chemical

PROCADIAN® UREA FEED MIXTURE

The quick source of protein in mixed feeds

Research Specialties Co. Begins Construction

Construction has begun on a new $275,000 manufacturing plant at Richmond, Calif., for Research Specialities Co., manufacturers of research instruments and radiochemicals for medical and industrial research.



The new plant will be devoted to the production of the company's line of radiochemicals, such as C^{14}-labeled compounds, and its research instruments.

L. K. Crowe Honored at Nebraska

L. K. CROWE, professor of dairy husbandry, Univ. of Nebraska, Lincoln, was honored recently as one of the 1958 recipients of the Univ. of Nebraska Foundation awards for distinguished teaching.

The awards, which consist of a $1,000 stipend and a medallion, were presented at the 30th Annual Honors Convocation by W. W. PUTNEY of Lincoln, past-president of the Foundation.

Dr. Crowe has been a member of the University staff since 1925. In nominating him for the Award, his colleagues said, "He has maintained a critical attitude toward his students' accomplishments, not being satisfied with less than the best, but tolerant of mistakes beyond control or mistakes not made too often. He has maintained a hearty laugh and a sense of humor, but at no time was he willing to sacrifice principles for the sake of popularity."

During his long career at Nebraska, Dr. Crowe has been a junior division adviser, sponsor of the Varsity Dairy Club, faculty representative of the Agriculture Religious Council, fraternity adviser for Alpha Gamma Sigma, and chairman of the University Wesley Foundation.

Cost Clinic Held at Georgia

The Southern Cost Analysis Clinic on dairy plant production, sales, and distribution was held in May at the Continuing Education Center, Univ. of Georgia, Athens. Representatives from nine states attended. Keynote speaker was S. G. CHANDLER of the Georgia Agricultural

THEY'VE BEEN A GOOD

53

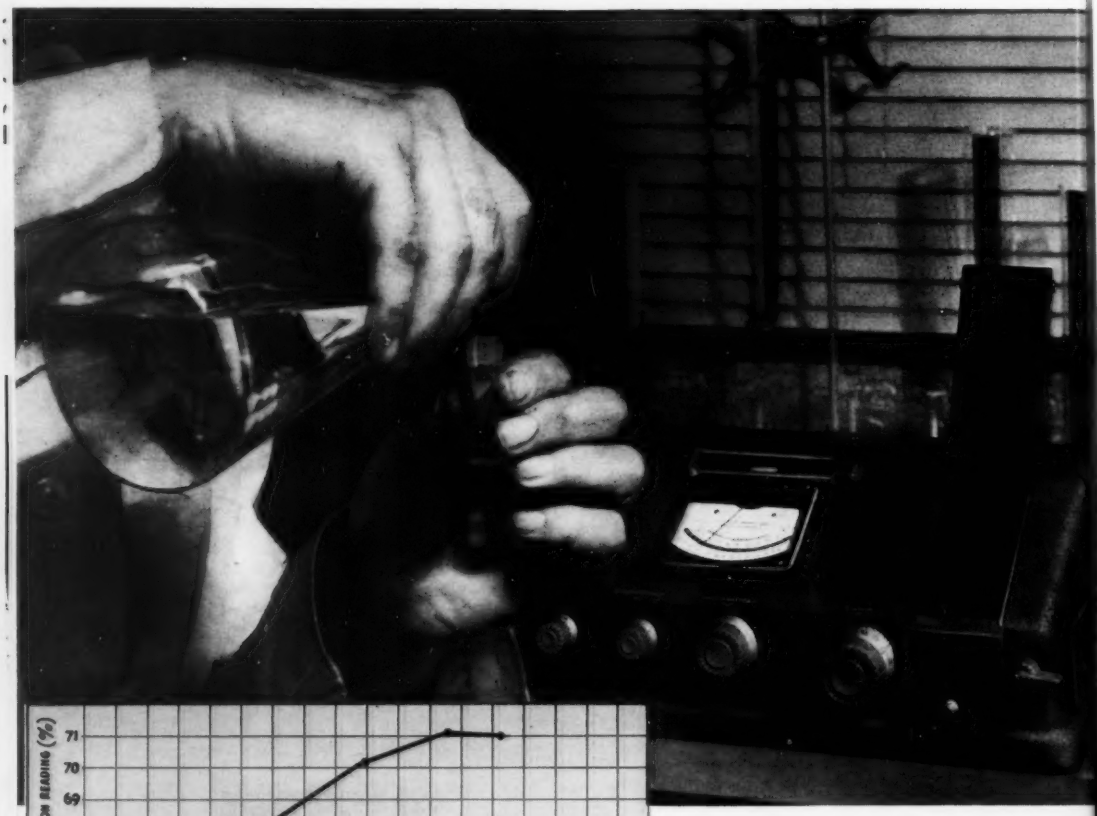
AMERICAN DAIRY YEARS, AND A LOT OF THEIR GOODNESS STEMS DIRECTLY FROM THE CONTRIBUTIONS OF AMERICAN DAIRY SCIENCE ASSOCIATION.
WE DAIRY INDUSTRIAL SUPPLIERS-EQUIPPERS ARE CONSCIOUS OF THE MAGNITUDE OF THE DEBT OWED BY THE DAIRY WORLD TO OUR 53-YEAR YOUNG SISTER ASSOCIATION.
WE'VE BEEN PROUD TO WORK CLOSELY WITH ADSA SINCE 1930 AS THE CO-SPONSOR OF THE COLLEGIATE STUDENTS' INTERNATIONAL CONTEST IN JUDGING DAIRY PRODUCTS.
WE'VE ALSO RECOGNIZED IT AS A PRIVILEGE TO WELCOME ADSA MEMBERS TO THE DAIRY INDUSTRIES EXPOSITIONS, WHICH WE'VE SPONSORED SINCE ORIGINATING THEM IN 1926.
(DON'T FORGET THIS YEAR'S BIG SHOW ON NAVY PIER IN CHICAGO, DECEMBER 8-13.)

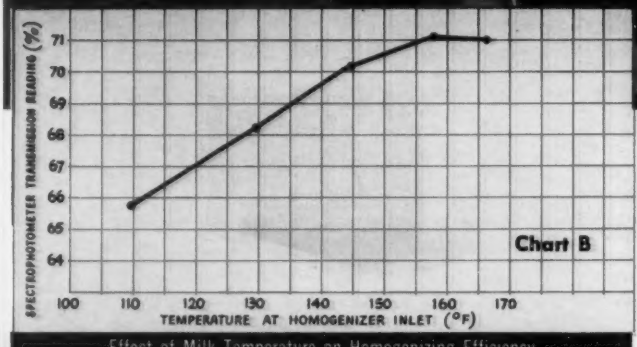
"MEET THE CHALLENGES"
DAIRY INDUSTRIES EXPOSITION
Navy Pier, Chicago
December 8-13, 1958

MORE GOOD YEARS STRETCH AHEAD . . . MAY THEY SEE EVER CLOSER AND MORE FRUITFUL COLLABORATION BETWEEN ADSA AND THE MEMBERS OF . . .

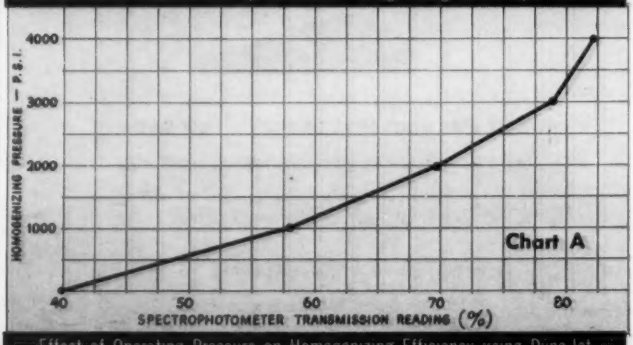
. . . Dairy Industries Supply Association
1145 19th Street NW, Washington 6, D.C.

There's a New Science





Effect of Milk Temperature on Homogenizing Efficiency



Effect of Operating Pressure on Homogenizing Efficiency using Dyna-Jet

Manton-Gaulin has compiled an impressive amount of data on efficiency of homogenization with this Spectro-photometer. Using light transmission as a norm, it is possible to precisely measure the degree of fat particle break-up produced by any given valve. The findings have been used to develop the new Dyna-Jet valve which produces a finer product, and sets new standards in homogenizing efficiency.

Gaulin Research Charts show how homogenizing efficiency can be measured by the Spectrophotometer method. Chart A illustrates the remarkable efficiency of Dyna-Jet and also the significant difference in the quality of the end product as operating pressures are increased. At any pressure, Dyna-Jet outperforms all competitive valves. Chart B shows the effect of milk temperature on homogenizing efficiency. Manton-Gaulin laboratory technicians will be happy to work with you on your homogenizing problems.

to Homogenizing!

Here's a completely different concept in looking at homogenizing efficiency. Years of study in the Manton-Gaulin Research Laboratory have produced a penetrating analysis of fat particle break-up and uniformity of product — and with it a revolutionary homogenizing valve. The most significant advance in a decade, dairymen can now look for a new degree of perfection in the homogenizing of milk.

This new science has been sparked by developing special light transmission tests using the Spectrophotometer technique, together with exhaustive compilation of comparative data. The result: For the first time, accurate, *visible* evidence on ability of homogenizing valves to produce finer and more uniform fat particle break-up, and a more perfectly homogenized milk. Truly, a new dimension has been added to dairy science!

The Dyna-Jet was developed by Manton-Gaulin to bring the results of this new science right into your own plant. Now, as never before, you can achieve perfection in homogenized milk — more digestible, tastier, richer in appearance.

Gaulin Dyna-Jet Homogenizers are here, now. Check the condensed story presented on these pages — then take steps to learn all the facts about this development. Perhaps you can be the first to capitalize on new Gaulin Dyna-Jet Homogenizing in your market area. For special literature and technical data, or for a personal presentation, contact your local Manton-Gaulin dealer, or write: The Manton-Gaulin Manufacturing Co., Inc., 44 Garden St., Everett 49, Massachusetts.

Gaulin TRADE MARK

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MANUFACTURING CO., INC.
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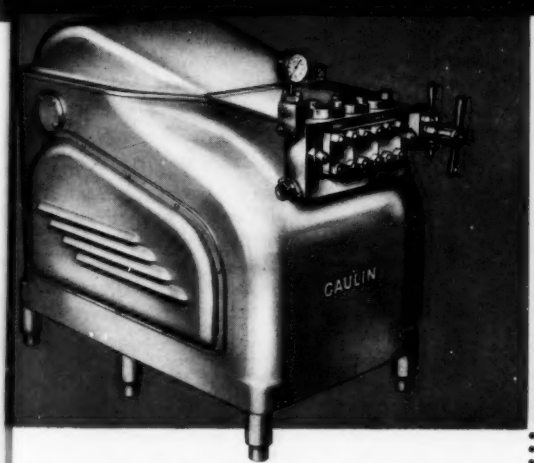
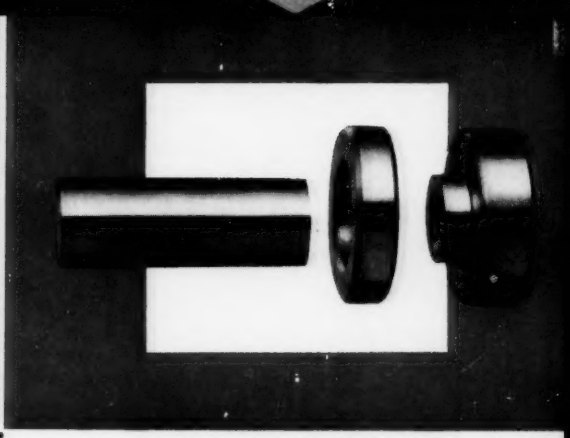
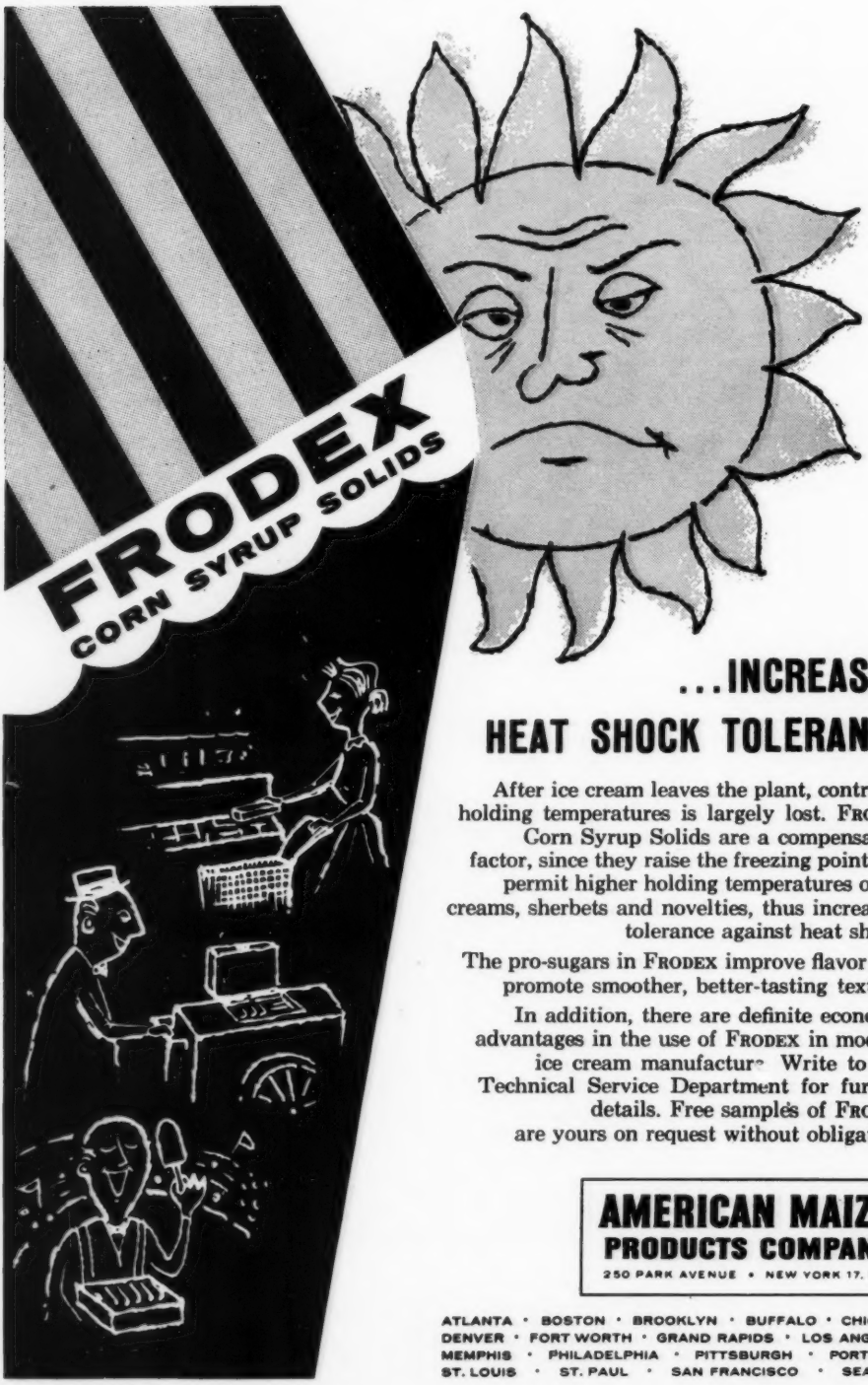
ld's largest manufacturers of homogenizers, colloid mills and triplex high pressure pumps.



This Gaulin DJ-40 is one of the new Gaulin Homogenizers designed to use the revolutionary Dyna-Jet valve. It retains the traditional Gaulin long-life construction and "floating-oil" features and achieves an even greater degree of homogenizing efficiency by using the Dyna-Jet.



This Exclusive Gaulin Dyna-Jet Valve provides a completely new concept in homogenizing. Dyna-Jet uses hydrokinetic principle, and is designed to create tremendous energy and velocity gradients, together with zones of extreme turbulence, to produce extremely fine fat particle break-up.



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After ice cream leaves the plant, control of holding temperatures is largely lost. FRODEX Corn Syrup Solids are a compensating factor, since they raise the freezing point and permit higher holding temperatures of ice creams, sherbets and novelties, thus increasing tolerance against heat shock.

The pro-sugars in FRODEX improve flavor and promote smoother, better-tasting texture.

In addition, there are definite economic advantages in the use of FRODEX in modern ice cream manufactur- Write to our Technical Service Department for further details. Free samples of FRODEX are yours on request without obligation.

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Extension Service. His topic was "Let the Cream Come to the Top."

Members of the Clinic faculty included E. C. BAKER, Foremost Dairies, Jacksonville, Fla.; W. J. BOOKHOLD and R. H. ELROD, Internal Revenue Service, Atlanta, Ga.; F. A. BUCHANAN and L. H. CASSMEYER, Pet Dairy Products Co., Johnson City, Tenn.; J. L. CAMPBELL, Dairy Sales Consultant, Dothan, Ala.; W. J. BORBETT, Dean Milk Co., Rockford, Ill.; A. C. FAY, Dairy Consultant, Miami, Fla.; J. E. GATES and H. M. HECKMAN, College of Business Administration, Univ. of Georgia; MARK HORNBUCKLE, Atlanta, Ga., Dairies; MARJORIE G. HUGO, Dairy Accounting Service, Lexington N. C.; N. KOHL, Southern Dairies, Atlanta, and BEN STAKES, Southern Dairies, Birmingham, Ala.

Kentucky News

Twenty-four students completed the spring session of the Artificial Breeding Short Course of the Univ. of Kentucky, Lexington.

The short course, which is presented twice a year, consisted of the practice and theory of artificial breeding. E. C. TROUTMAN was chairman of the course.

A recently held Dairy Career Day at the Univ. of Kentucky attracted about 70 high school boys. The pupils were sponsored and accompanied by representatives of dairy organizations throughout the state.

The boys were shown the facilities for work in dairy production and manufacturing in the University's laboratories. Opportunities of the industry were discussed by MARVIN GUTHRIE, Pet Milk Company, and ROBERT JOHNSON, Oscar Ewing, Inc. W. A. WENTWORTH, former vice-president of the Borden Company, also discussed the need for trained men in all phases of the dairy industry.

Those attending the Career Day were guests of the Kentucky Dairy Science Club members at their annual Honor Banquet, where JOHN KLOECKER, Lexington, was named honor guest.

Cattle Sale in Louisiana

The Louisiana Holstein-Friesian Association held its sixth annual breed promotional sale at Baton Rouge in May.

Thirty-eight head of bred and open heifers, consigned by the Fox River Valley Holstein Breeders Association, Fond du Lac, Wis., brought an average of $412. The top price of $705 was paid for Wis Reva Symbol, a 24-month-old bred heifer, by Burl Roy of Shreveport, La. Ten of these heifers were under one year of age; the rest were senior yearlings and 2-year-olds.

KLENZADE
SODIUM HYPOCHLORITE
SANITIZERS

SAFE for finest stainless steel surfaces. 25 years of daily use in thousands of plants prove it *conclusively*.

Klenzade X-4, Klenzade XY-12, and Klenzade Tri-Chloro-Cide have the *highest* germicidal power of all chlorine-bearing sanitizers on the market. Comparative test data on request.

Most economical, too. Always ready for use . . . no "settling out" . . . no residual film . . . exceptionally long in-plant keeping quality.

Service Throughout America

KLENZADE PRODUCTS, INC.
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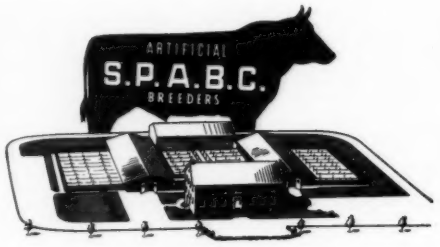
Pennsylvania's Modern Dairymen . . . .



PLAN . . . .

SELECT . . . .

PRODUCE . . . .

Dairy Herd Improvement throughout the "Keystone" State today commands profound recognition. We here in Southeastern Pennsylvania are contributing toward such success by availing the services of "Sires of Prominence." The S.P.A.B.C. Proved Sires are all type and production accepted and approved. Results of this sound program are being amplified in the herds of our many members.

Science and Research continues its pace by contributing toward our search for ways to improve our methods of operation and handling. None the least, we are deeply appreciative to Dr. J. O. Almquist and his workers at Penn State for their "diluter discovery" whereby we are permitted to fully utilize fresh supplies on an "Every Sire, Every Day" plan. This, we are happy to report, is being made available to our members at an added economy and with a higher rate of breeding efficiency.

We at S.P.A.B.C. proudly announce that 28.8% of our 1957 services were performed on Registered Cows in the five dairy breeds. Suffice to say, the constructive influence of this program is being indelibly written into the record of today's purebred industry. We repeat our aim, "Better Cattle For Better Living."

Whether it's OLD (still the same $5.00 service fee) or NEW (choose the sire of your choice), we stand ready to "Ring The Bell' for you. If you live in Southeastern Pennsylvania or the State of Delaware, ask your neighbor and he will assure you of our SERVICE, SIRES and ECONOMY.

SOUTHEASTERN PENNA. ARTIFICIAL BREEDING COOPERATIVE

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Minnesota Dairyman Receives UDSA Award

RALPH WAYNE, extension dairyman at the Univ. of Minnesota, St. Paul, has received the Superior Service award from the U.S. Department of Agriculture. This award is the highest honor the UDSA can give one of its employees.

Wayne was recognized for his outstanding record as an educator and for organizing, developing, and carrying through several "pioneer" programs of benefit to Minnesota people.

In one of his most recent projects, Wayne led the educational work in eradicating brucellosis from Minnesota cattle herds. He worked closely with the State Livestock Sanitary board, the U.S. Department of Agriculture, country agricultural agents, and farmers in this program. Minnesota received "modified-certified" brucellosis-free status in May, 1957. It was the seventh state to do so.

Wayne was born and reared on a Freeborn County farm. He attended Carleton College in 1924–25, spent a year and a half working on farms, then entered the Univ. of Minnesota in 1927 and received his B.S. "with distinction" in 1929. In 1931, he earned a M.S. degree in husbandry and agricultural biochemistry.

He spent the 1931–32 academic year at the Royal Veterinary and Agricultural College in Copenhagen, Denmark, and in studying agriculture in eight other countries. For the next ten years, he was agricultural agent at Litchfield, in Meeker County, and from 1942–45 was public relations director and worked in production development for Land O'Lakes Creameries, Inc.

He took his present position in 1945 and during part of the next year took a leave of absence to serve as agricultural representative for the U.S. Foreign Economic Administration mission to Denmark.

He is a member of Alpha Zeta, Gamma Sigma Delta, Gamma Alpha, and Epsilion Sigma Phi, national honorary fraternities. He is a Fellow of the American-Scandinavian Foundation and is a life member of the Royal Agricultural Society of Denmark.

Projects, Consultation, and Production Control Services in

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Write for price schedule.

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IODINE SANITIZERS
OFFER ALL THESE ADVANTAGES

A LONG RECORD OF DEPENDABILITY. Iodine is recognized as a most efficient antiseptic and germicide. It is known to be effective against a wide range of organisms. New technology has now resulted in more efficient iodine formulations developed especially for sanitization.

EASY TO USE. Iodine sanitizers are formulated especially for treatment of dairy utensils and equipment. Leading manufacturers offer iodine sanitizers and detergent-sanitizers as liquids, powders or tablets.

EFFECTIVE. Iodine sanitizers are effective in low concentrations . . . economical, too. They can help you supply better milk.

EASY TO TEST. The well-known iodine color is an indication of solution strength. When the color of an iodine sanitizing solution begins to disappear, that is a signal to replenish or replace the solution. Test kits are available.

Write us for further information and names of manufacturers offering iodine sanitizers in your area. No obligation, of course.

CHILEAN IODINE EDUCATIONAL BUREAU, INC.

quality-control at the high concentrators

Fresh, rich milk from inspected farms . . . processed in clean, modern plants located throughout Wisconsin . . . inspected, tested again and again, at every step in production — guarantees the uniformly high quality of Morning Glory nonfat dry milk solids. In the development and application of this product, Consolidated Badger has shared a productive partnership with the dairy scientists of America.

Morning Glory
NONFAT DRY MILK SOLIDS

CONSOLIDATED BADGER, INC., SHAWANO, WISCONSIN

STUDENT CHAPTER NEWS

J. E. JOHNSTON, *Editor*

Connecticut Dairy Club News

The Connecticut Dairy Club activities for the current year started with the Annual Dairy Club Supper. The supper was followed by a talk by R. Niederman, illustrated with slides of his trip through the western United States last summer. The next major activity was the sponsoring of the Annual Dairy Cattle and Dairy Products Judging Contest, in which all interested students could participate.

The Dairy Products Judging Team placed first in the National Dairy Products Judging Contest, held at San Francisco, California. Team members are: E. Fleuette, top individual in the nation, E. Geishecker, and P. Schoek. The team also took first honors at the Eastern States Exposition held at West Springfield, Mass.; the team members were the top three men in the contest.

Subjects presented at the biweekly club meetings have included: "The Artificial Breeding Program," by J. Helm, Field Service Representative of the Connecticut Artificial Breeding Association; "Milk Co-operatives," by K. Geyer, manager of the Connecticut Milk Producers Association; "Half-Gallon Container Issue," by L. Beebe, owner of Beebe's Dairy in Norwich. Other subjects have been: "Changing Times and Changing Practices in Dairying," by R. Hurlbutt, a Pomfret dairy farmer and UConn graduate of 1951; "The Vacuum Treatment of Milk," by R. Soderlund of the DeLaval Separator Company; "Mastitis Control," by H. Hale, Assistant Professor of Animal Diseases at UConn. Dr. R. Cleverdon, Professor of Bacteriology, gave an enlightening and entertaining talk concerning "Milk Control Systems of the Near Future," which was illustrated with many humorous anecdotes. "Experiences in Dairy Farming," by W. Wadsworth, owner of a large dairy farm in Farmington, and "The Importance of Marketing in the Dairy Industry," by C. Brock of Brock-Hall Dairy of Hamden, were the last two subjects dealt with.

Rutgers Dairy Science Club

The Rutgers Dairy Science Club has been quite active during the past few months.

Beginning in the fall with the Ag Barn Hop, which is meant to help relieve frustrated students from the "studying grind," the Dairy Club entered an exhibit, the theme being, "I've Got A Secret." The secret was that "Even The Greatest Scientists Don't Know How I (The Cow) Change Grass into Milk." Club members were well pleased with the completed exhibit.

The Fitting and Showmanship Contest, sponsored by the Dairy Club and held between the short course and four-year students, took place the following month, December. Strange as it may seem, the contest ended in a draw. Consequently, the coveted Bartlett Trophy was presented to a representative of each team. We are proud to say that the president of the Dairy Club, John Teller, was the over-all winner in showmanship.

The next major event on the calendar for the Dairy Club was the annual luncheon which the club prepared for a conference of men in the dairy industry, held on the Rutgers campus. The luncheons are not only convenient for those attending the conferences, but they are also a source of income for our treasury. This year we cleared $139.50.

At the present time, the Club really has its hands full. We are working on the Dairy Club Yearbook and hope to have it at the printers within a week. We are also looking forward to Ag Field Day, which is the climax of competition for the various agricultural clubs on campus. Winding up our program for this year will be a picnic held at the rustic Log Cabin on the ag campus at Rutgers, an event which is always enjoyed by all.

Purdue Dairy Club Report

Approximately 115 members and guests attended the annual Dairy Club Banquet, held at 6 P.M. in the new Home Economics Building cafeteria, April 29. The toastmaster for the evening was the club's president, Don Wilkey of Farm House Fraternity.

The banquet was started with a cheese smorgasbord, in which were almost forty different varieties of cheese. The display was very colorful as well as appetizing. The rest of the banquet was eaten and finished with ice cream fruit molds as dessert. Guests were introduced, which included Ed Gannon, a donor of some of the awards, Professor Pfendler, Assistant Dean of Agriculture, and Mrs. Pfendler, and Professor Gregory, who is the head of the Dairy Department, and Mrs. Gregory.

Entertainment for the evening was furnished by Don Burns, who is an amateur hypnotist. Although he did not have very good subjects, he did give an informative and interesting demonstration of the powers of hypnosis.

The last item on the program was the presentation of awards for the production and manufacturing contests. These awards were presented to the winners of these contests by our faculty sponsors, Professor B. V. Crowl of the Production Department, and Professor D. R. Frazeur of the Manufacturing Department. Over-all winners of the production classes of

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TWIN-PAK

Safer...

3 Vital Health Ways

1. Safer When She Buys
2. Safer When She Uses
3. Safer When She Stores

And in addition . . . the housewife knows that the NEW Sealright TWIN-PAK is Wax-free, Leak-free, Waste-free and always gently pours.

"Pour One, Store One" is a merchandising refrain you'll hear housewives countrywide repeating; it means TWIN-PAK, the NEW, NO-WAX milk container is the homemakers approved modern way to package, use and store milk.

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Sealright

Sealright-Oswego Falls Corporation, Fulton, N. Y.—Kansas City, Kansas—Sealright Pacific Ltd., Los Angeles, California—Canadian Sealright Co., Ltd., Peterborough, Ontario, Canada.

all breeds were: First, Bruce Wiley; second, Rolland Kline; third, Tom Dunlap; fourth, Loyd Young; fifth, Charles Bastian. The best set of reasons on all breeds was given by Tom Dunlap. The sophomore class was chosen as the class with the best over-all judging. These members were Loyd Young, Charles Bastian, and Art Strong.

In the manufacturing contest, the first-place winners were: butter, Dale Wolfe; cheese, Fred George; ice cream, Raymond Brinegar; milk, Clifford Gerbig. The first-place individual judge of all products ended in a tie between Fred George and Clifford Gerbig. This was the first time in ten years that there has been a tie for this award. The class winner was the senior class—Fred George, Clifford Gerbig, and William Daugherty.

The officers of the club, who are Don Wilkey, president; Larry Stallwood, vice-president; Bob Ulrich, secretary; Norris Barnett, treasurer; and Tom Swinford, publicity, were recognized for their efforts in making the banquet a success.

L.S.U. Dairy Science Club News

On Saturday, March 27, the Future Farmers of America in the Baton Rouge area held a judging contest in the Agricultural Center show barn. This is an annual event sponsored by the L.S.U. Dairy Science Club. The Dairy Judging team does the greatest portion of the supervision necessary in the contest. These boys are assisted by club members and faculty of the Dairy Department. There were several classes of livestock, and the F.F.A. had an interesting and educational experience. In addition to this contest, the club sponsors district and state F.F.A. and 4-H judging contests.

On Saturday, April 12, the members of the club held their annual dairy judging contest. This contest included both dairy products and dairy animals. The members were divided for each contest into senior class (juniors and seniors) and junior class (freshmen and sophomores) judging teams. Prizes were awarded for each division at the annual spring barbecue held on April 19 after the faculty-student baseball game. The students won this affair again, but only after a tough battle.

The final event on the social calendar of the club was the annual banquet and dance held May 3. In attendance were 112 club members, alumni, faculty members, and women guests. The coveted "Dairyman of the Year" award was made to Mr. Bert Davis, owner of the Milkhouse Dairy, Winnsboro, Louisiana, in recognition of the Dairy Industry of the state. Mr. Davis is an alumnus of the club and has done much to promote the growth of our dairy industry. Cur-

Ali Baba *and the* 40 *Thieves*

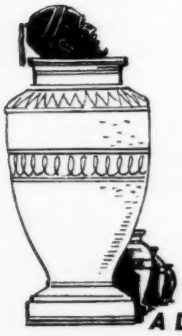
The fabled Ali Baba never suspected that he was harboring 40 armed thieves under his roof. The fable states that Ali's watchful servant, Morgiana, discovered them lurking there and saved Ali and his valuables.

There are 40 thieves lurking in every business operation today — awaiting every opportunity to rob it of profit. There are Wastes, Production Losses, Distribution Costs and a host of others — most of them well entrenched and not easily recognized.

That's why every business needs a watchful servant like Morgiana — a faithful guardian of profit. In the Dairy Industry that has been the function of the Dairy Manufacture and Research Bureau for 30 years

The Bureau uses a sharp and practised eye for ferreting out of Milk and Ice Cream operations the 40-odd leaks that may be robbing them of hard-won profit.

If you suspect **you** are paying tribute to some of these hidden thieves, why not join the Bureau **now**? Avail yourself of the Bureau's many services which will put your operation back on the road to **full** profit. For complete information, write



The Dairy Manufacture and Research Bureau

A Division of **G. P. Gundlach & Company**

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Report | from Carnation Research Laboratory

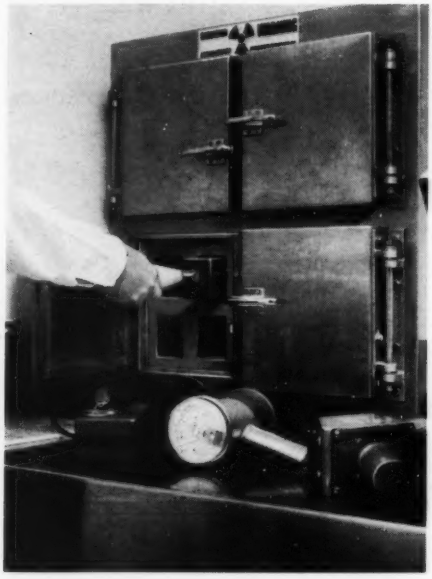
Van Nuys, California

5-Phase Research

For a half century, Carnation has conducted a continuous and expanding 5-phase research program in dairy and cereal products.

1. CARNATION FARMS

More than fifty years of scientific cattle breeding at Carnation Farms have resulted in many famous Holstein champions. Cattle from these famous bloodlines are shipped to dairy farms across the country to help improve the Carnation milk supply.



2. CARNATION RESEARCH LABORATORY

Newest research facility is the Carnation Laboratory at Van Nuys, California, shown above–one of America's most modern laboratories devoted to product research. In addition to direct research in dairy and cereal foods, Carnation Laboratory coordinates the other phases of Carnation Research.

3. QUALITY CONTROL RESEARCH

Laboratories at each Carnation Plant exercise rigid day-by-day control of Carnation Milk processing. Samples from *all* Carnation plants are rechecked at Central Product Control, Oconomowoc, Wisconsin, to assure *uniform* high quality of Carnation Milk everywhere.

4. SPONSORED ASSOCIATION RESEARCH

As a member of the National Research Council, Evaporated Milk Association, National Dairy Council, American Dry Milk Institute and many other similar organizations throughout the world, Carnation contributes importantly to the broad research activities conducted by these groups.

5. SPONSORED UNIVERSITY RESEARCH

Carnation sponsors University research in highly specialized fields, such as radiation and supersonic vibration, as related to dairy foods. Current projects are under way at the California and Massachusetts Institutes of Technology and the Universities of Wisconsin and Illinois.

CARNATION QUALITY FOODS ARE PROTECTED BY CONTINUOUS 5-PHASE RESEARCH:

Carnation Farms,
Carnation Research Laboratory,
Carnation Plant Laboratories,
Carnation Central Product Control Laboratory,
Carnation-sponsored University and Association Research.

"from Contented Cows"

rently, he is president of the Louisiana Dairy Products Association. Awards were also presented to Ken Conly, 1957–58 club president, for outstanding scholastic achievement and to Bruce Jones, our national champion dairy cattle judge, as outstanding senior. The banquet was followed by a dance, which was thoroughly enjoyed by all.

Dr. J. C. Knott in characteristic pose, at the dinner dance held in his honor.

Achievements of club members this year included the recognition of club president, Ken Conly, as outstanding senior in the college of agriculture at the spring convocations of the college and the university, as well as the election of Steve Conerly, club member, as president of the college of agriculture for the 1958–59 school year.

Washington State College News

The Washington State College Student Affiliate Chapter of the A.D.S.A. sponsored a dinner and dance on January 18, in honor of Dr. J. C. Knott. Joe, as he is affectionately known, is retiring at the end of this academic year, after serving the dairy and agricultural industries for nearly 40 years (see cut).

The students sponsored a "This Is Your Life" program. They were ably assisted by former teachers, classmates, and long-time friends of Dr. Knott. At the termination of the program, he was presented with a large volume of letters and telegrams from friends from all parts of the nation.

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In addition to supplying you with high quality chocolate flavorings and coatings for your ice cream, The Man From Blumenthal will gladly:

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2. Arrange for his laboratory to perform special services that can only be handled by a lab staffed with chocolate technicians and equipped with the latest analytical and pilot plant equipment to produce special types of chocolate for ice cream.
3. Refer your problems to one of our consultants specializing in technical ice cream problems.
4. Conduct research on new ideas suggested by you.

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3. **To provide a reliable Every Cow Check-up for mastitis, improved milk quality and lower production costs.**

RAPID-FLO® Single Faced Filter Disks are engineered by Johnson & Johnson to provide safe filtration and the all important RAPID-FLO® Check-up for mastitis and extraneous matter. When you recommend engineered RAPID-FLO® Single Faced Disks and the RAPID-FLO® Check-up, you are helping the producer improve milk quality and increase profit.

Johnson & Johnson **Filter Products Division**

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Cornell Club News

The Cornell Student Chapter of the A.D.S.A. held its annual banquet at the Lehigh Valley House on Saturday night, May 10.

A Special Distinguished Service Award was presented to Dr. E. S. Guthrie, professor emeritus, for fifty years of continuous service to Cornell University and the dairy industry of New York State. The Award cited, in particular, his guidance and counsel of students in personal, academic, and club matters through the many years of his service. Dr. Guthrie came to Cornell in 1908 as a graduate assistant. He received his Ph.D. in 1913 and was appointed an Instructor in Dairy Industry. In 1918, he was promoted to Professor of Dairy Industry, in which capacity he served for 30 years. In 1948 he retired officially, but unofficially has been as active as ever, and has continued to conduct research along the lines of his major interest—the flavor quality of milk. Dr. Guthrie is a charter member of the American Dairy Science Association and has held many offices in this organization, including the vice-presidency and presidency, in 1939 and 1940, respectively. He was awarded an Honorary Life Membership in 1944.

Each year, the Cornell Dairy Science Club presents a Dairy Industry Award to an outstanding leader in the dairy industry in recognition of his lasting contributions to the dairy industry in New York State. This year the honor was extended to Henry W. Lehmkuhl, President

Leading dairymen say . . .

WE HELPED

All of us within and alongside the dairy industry rejoiced when recent figures indicated the average cow has passed the 6,000 lb. per year production mark.

It could and should be a lot higher in years to come.

We'd like to think we had a part in this progress. A recent nationwide survey including replies from 884 leading dairymen expresses this "attitude"——

Research in animal nutrition by leading feed companies has done much to increase milk production in the U. S.—

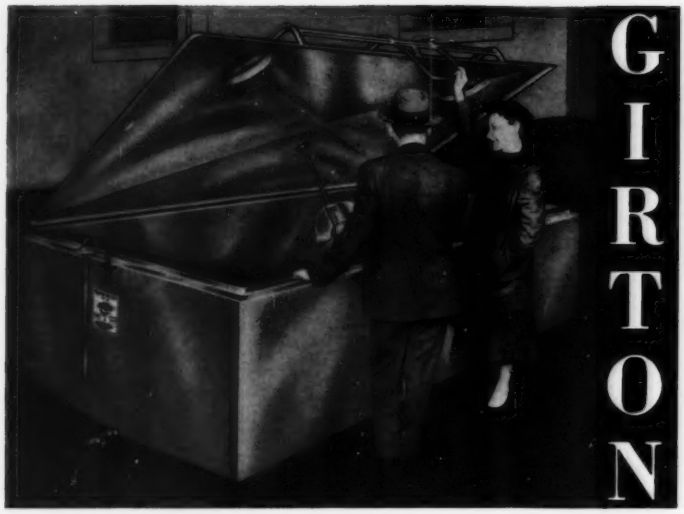
Agree very much	54.0%
Agree somewhat	35.5%
Disagree a little	4.8%
Disagree very much . . .	2.7%
No Answer	3.0%

Certainly, our research has improved our product. But, if it also helped the industry, we are prouder still.

May our research team . . . along with yours . . . continue to improve the industry we serve!

RALSTON PURINA COMPANY

St. Louis, Mo.



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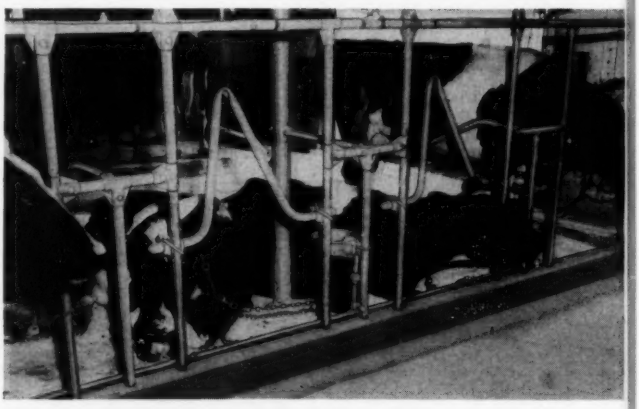
INTRODUCING THE NEW ONE-PIECE COVER MODEL

So EASY to POUR INTO and so EASY to CLEAN

GIRTON Dairy Equipment

THE CONTROL OF THE STANCHION STALL, THE CLEANLINESS OF THE TIE STALL AND THE FREEDOM OF THE CONVENTIONAL COMFORT STALL ARE THE FEATURES INCLUDED IN



The GIRTON COMFORTMASTER STALL

GIRTON MANUFACTURING COMPANY
• MILLVILLE PA.

of Milk Plant Specialties Corp. in Rochester, New York. He graduated from Columbia University with a degree in Pharmaceutical Chemistry and soon after held the position of chief of the Division of Food and Drug Inspection of the New York City Department of Health. Mr. Lehmkuhl has operated his own technical laboratories and provided cleaners and sanitizers to the dairy industry in New York. In this capacity, he has been of considerable help to dairy plants and dairy farmers. In 1930, he was called to New York City at the recommendation of the Millbank Foundation to reorganize the Food and Drug Department while serving as Director and Consultant. He has been extremely active in the New York State Association of Milk Sanitarians, as a member of their executive committee for six years and president of the Association in 1952. He has also been very active in the Rochester Area Sanitarians Association and a member of many of the affiliate sanitarian associations in New York State. Since 1955, he has served as President of the Council of Affiliates of the New York State Association, an organization established to help milk and food inspectors and the dairy plants' fieldmen with their "grass root" problems.

The Club also presented a key and monetary award to one of their members, Richard Scanlon, a sophomore in Dairy Industry, for his outstanding interest and service to the club this year.

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Reliable
HIGH QUALITY SINCE 1878
★ Cheese Rennet and Color
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A successful milling operation is dependent upon thorough knowledge of nutrition, unquestionable product quality, rigid control to standards and constant research. Recognize the similarity between these qualifications and those required to produce vitamin products? Twenty five years ago General Mills started supplying vitamin concentrates to dairies. Today dairies throughout the United States are users of General Mills Vitamin Concentrates.

General Mills Vitamin Concentrates give you these advantages:

1. A complete line of vitamin concentrates to choose from—to enable you to produce Vitamin D, Vitamin A & D, and Multi-Vitamin Mineral Milk.
2. Tailormade for any size operation.
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4. Package promotions created by General Mills advertising agency to help dairies sell their various types of milk.

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FEED COMPOUND

has a protein equivalent of 262%

One hundred pounds of Du Pont "Two-Sixty-Two" contains as much nitrogen as 262 pounds of protein. That's the amount of protein in 660 pounds of 40% oil meal feeds.

This highly-concentrated, readily-available nitrogen in "Two-Sixty-Two" nourishes and stimulates rumen bacteria—helps them multiply and develop faster. As a result, billions of these organisms are better able to help dairy cows break down and digest coarse fibers thoroughly and efficiently.

And this highly-concentrated urea source of protein in Du Pont "Two-Sixty-Two" also makes it possible for feed manufacturers to "concentrate" their protein and include more carbohydrates, minerals and vitamins in dairy rations. "Two-Sixty-Two" is used by hundreds of feed manufacturers in both complete feeds and concentrates.

These many advantages of "Two-Sixty-Two"—the first and finest urea source of protein—are the result of extensive research investigations by Du Pont and others in the interests of the dairy industry.

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Polychemicals Department
Wilmington 98, Delaware

Ralph D. Peterson, a junior from Jamestown, N. Y., was given a check for $100 for an essay entitled "Increase Milk Sales with Quality." Joseph C. Langberg, a senior from Peterboro, N. Y., received the second prize and a $25 check for his essay entitled, "Improve the Taste of Milk." Both students are majoring in dairy industry. The essay contest is open to everyone in the College of Agriculture. The contest developed from an endowment given to the College of Agriculture by Samuel L. Stewart to promote the production and distribution of high-quality milk.

The Dairy Science Club installed its officers for the coming year. They are Dominick V. Fonteechio, Amsterdam, president; Ralph D. Peterson, Jamestown, vice-president; Richard A. Scanlan, Altamont, secretary, and Paul M. Renckens, Dunkirk, treasurer. The retiring officers are Donald L. Henry, president; William J. Ernst, Jr., vice-president; William J. Herz, secretary, and Robert H. Johnson, treasurer.

Kentucky Dairy Club News

JOHN H. KLOECKER, president of Dixie Ice Cream Company, and secretary-treasurer of Ashland Dairy, Lexington, Kentucky, was named honor guest at the Honor Banquet held by the Univ. of Kentucky Dairy Science Club.

FOR ICE CREAM SALES

Feature monthly specials of 13 wonderful flavors—simply ripple Balch's Ready-to-Use Ripple Sauces into your own vanilla ice cream. NOTE: All Fruit and Butterscotch Ripple sauces are PURE!—contain no artificial flavor.

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NOW IN USE IN 26 COUNTRIES!

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Pure-Pak paper milk cartons are a product of Ex-Cell-O Corporation, Detroit 32, Michigan

The award is given by the Club annually for outstanding accomplishments in the field of dairying at the national, state, and local level. John O. Barkman, extension specialist in dairying, in presenting the guest said, "Mr. Kloecker has been prominently identified at all of these levels over a period of many years."

Currently, Mr. Kloecker is serving as a director of the Dairy Products Association of Kentucky, and is a regional director of the National Independent Dairies Association. He has been a director of the International Ice Cream Association and president of the Lexington Chamber of Commerce, as well as holding a number of other civic and industrial positions.

RICHIE LOWE, president of the Dairy Science Club, presented a picture of the honor guest to H. B. PRICE, acting dean of the College of Agriculture. Dr. Price said the picture would be hung in the Dairy Products Building at the University.

Prizes were awarded to winners in the Intra-Collegiate judging contest. The following students received one or more prizes: GLEN GOEBEL, CHARLES CORNETT, HOWARD PEAKE, STEWART BERRYMAN, DON MOORE, RONALD JOHNSON, DON HEILMAN, LEROY MCMULLEN, KELLEY THOMPSON, RICHIE LOWE, HERBERT MCCOMAS, MAX WHITE, MILTON WILLIAMS, and JOE ARNEY.

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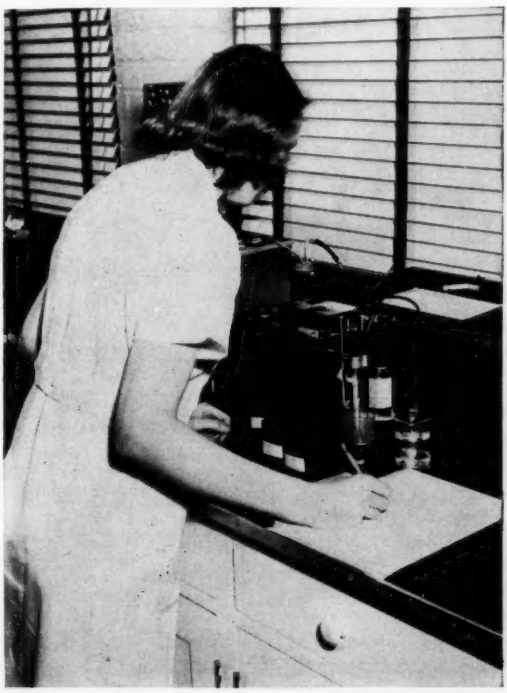

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We Specialize in IDEAS
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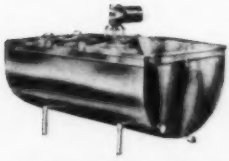

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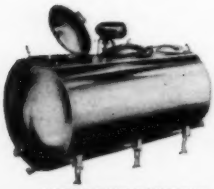
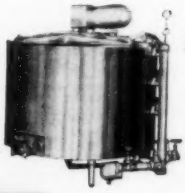
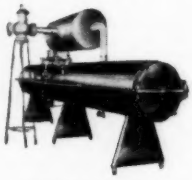


VACUUM BULK COOLERS



ZONE-CONTROL VATS



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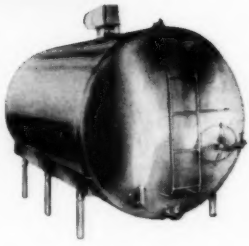

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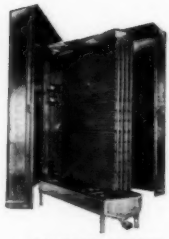

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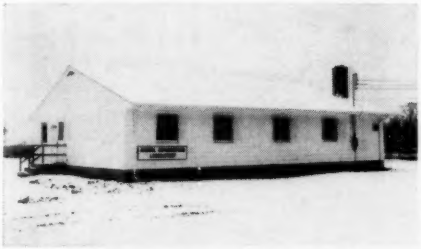

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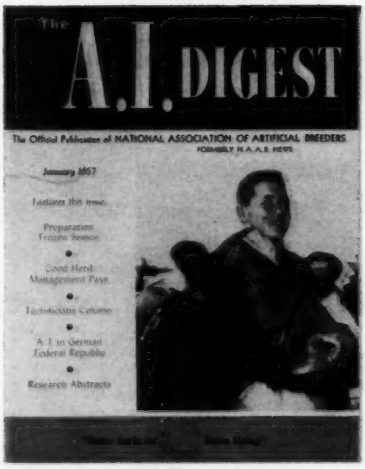

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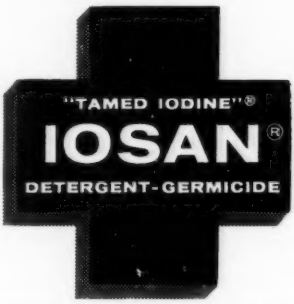

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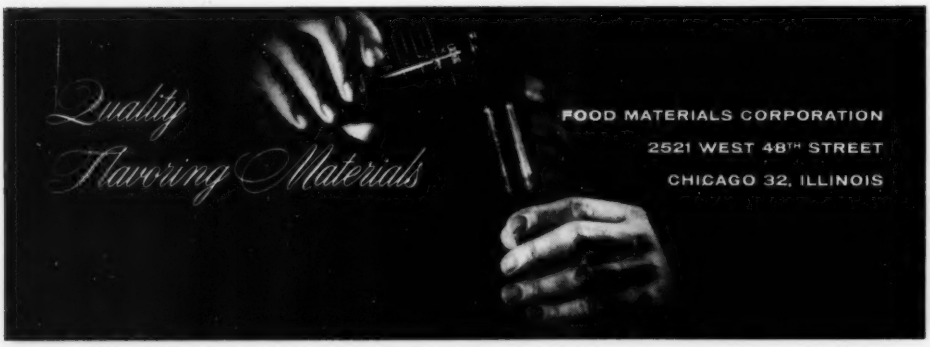

DISPERSIBILITY OF FOAM-DRIED FAT-CONTAINING MILK PRODUCTS

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Eastern Regional Research Laboratory,[1] Philadelphia 18, Pennsylvania

SUMMARY

It was found that foam-dried whole milk could be stored for at least 1 yr. at 73° F. without losing its excellent dispersibility. However, some loss was found when the material was stored at 100° F. Forewarming at 190° F. for 2, 5, and 8½ min. caused no significant change in initial dispersibility, but after storage at 100° F. for 1 mo. the dispersibiltiy was found to vary inversely with the holding time used in forewarming. The foam-drying process was successfully extended to a lactose-enriched whole milk, an acid-modified whole milk, and a chocolate-flavored whole milk. Products of enhanced dispersibilty were prepared by this method when compared to commercially available materials of similar types.

The dispersibility of nonfat milk powders has not yet been matched commercially with dry whole milk. The fat content of whole milk, amounting to a minimum of 26% by weight of the dry product, poses many problems not found with dry nonfat milk. A foam-dried whole milk *(3)* possessing excellent dispersibility in ice water with spoon stirring has been produced successfully on a pilot plant scale in the Philadelphia Laboratory of the Eastern Utilization Research and Development Division of the Agricultural Research Service. The current paper discusses the effect of storage on the dispersibility of the foam-dried product made from flash-pasteurized whole milk, with and without additional heat treatment of the whole milk. The value of foam drying as a means for improving the dispersibility of other dry fat-containing milks is discussed.

RETENTION OF DISPERSIBILITY ON STORAGE

Storage tests were made on foam-dried whole milk containing 29.5% fat and 2½% moisture, crushed to pass through a 20-mesh screen, and packaged in cans under an atmosphere containing 98.5% N_2 and 1.5% O_2. The cans were stored at 38, 73, and 100° F. and evaluated for dispersibility after storage periods of 2 wk. and 1, 2, 4, 6, and 12 mo. Dispersibility rate curves *(3)* were used to evaluate the effect of storage on the dispersing property. The tests showed that the dispersibility of foam-dried flash-pasteurized whole milk remains unimpaired for at least 1 yr. at room temperature (see Table 1).

The reasons for this good dispersibility and its permanence are not fully understood. However, some of the contributing factors are as follows:

1. *Avoidance of protein alteration during processing.* That the proteins are not denatured is evidenced by ultracentrifugal tests [described in *(3)*] which show the whey proteins to have the same sedimentation pattern as those in fresh pasteurized milk (see Figure 1). As might be expected, the

Received for publication November 7, 1957.

[1] A laboratory of the Eastern Utilization Research and Development Division, USDA.

TABLE 1

Effect of storage at 73° F. for one year on dispersibility rate of foam-dried whole milk

Stirring time	Dispersibility rates	
	Fresh	Stored
(sec.)		
20	70.2	76.4
40	87.8	84.3
60	93.7	93.6
80	95.3	94.5
100	95.3	96.8
120	97.8	98.1
Mean dispersibilities	90.0	90.6

product contains only trace amounts of undispersed material by the ADMI solubility index test.

2. *Dispersion of the fat globules which remain discrete throughout processing and storage.* Microscopic examination of the fat in the concentrate before drying, and in the milk reconstituted from the dried stored product, shows no evidence of coalescing or clustering. The fat globules remain discrete and are of the same size as immediately after homogenization. They appear to be "bound," perhaps by physical-chemical means, within a matrix of readily dispersible material.
3. *The distinctive shape of the particles.* Unlike spray-dried particles, particles of crushed foam-dried milk are irregular in shape, have one very small dimension, are not powdery; hence, have little tendency to cake- or ball-up in contact with water.

Storage at 100° F. impaired the dispersibility (as shown in Figure 2). After 1 mo. of storage there was a loss of about 5% in dispersibility with 2 min. of

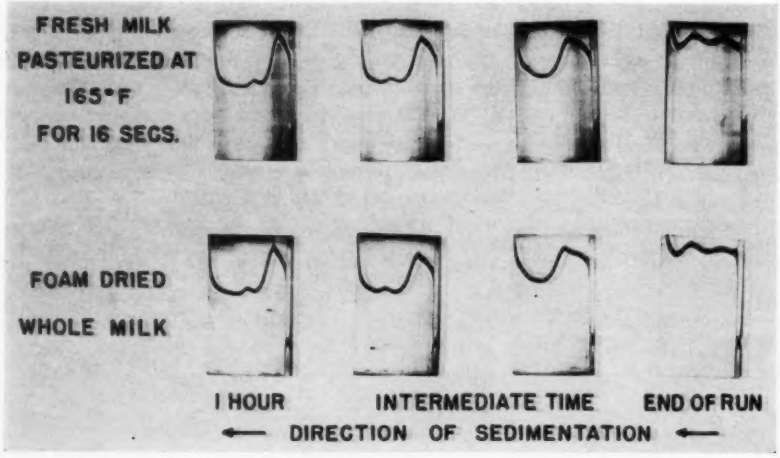

FIG. 1. Ultracentrifugal patterns of whey protein concentrates *(4)*.

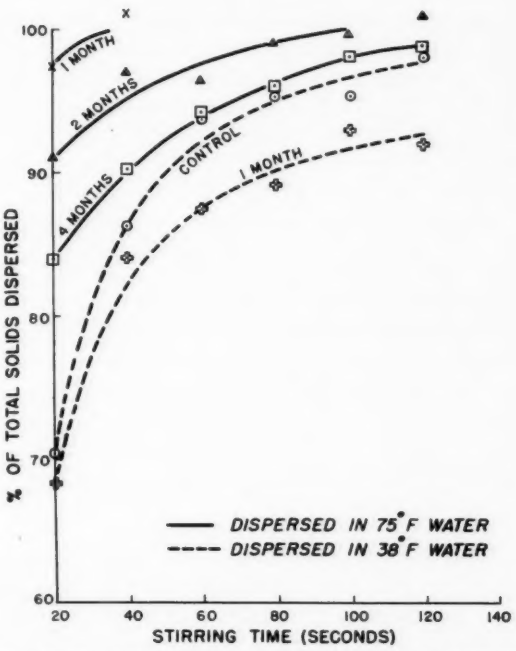

FIG. 2. Dispersibility rate changes with storage at 100° F.

stirring in 38° F. water. However, when 75° F. water was employed, which is the temperature normally used in dispersibility tests, complete dispersibility resulted after only 35 sec. of stirring. Even after 4 mo. of storage at 100° F., dispersibility in 75° F. water was excellent.

Microscopic examinations of the fresh and stored samples showed in both cases that the fat globules were imbedded in a milk solids-not-fat matrix. The only apparent change resulting from the storage was in this matrix. In the case of the stored sample, it disintegrated slowly in water. There was no evidence, however, of "fat migration," to which the well-known loss of dispersibility in some dry whole milks has been attributed. It appears probable that the loss in dispersibility is a consequence of protein insolubilization resulting from high-temperature storage.

EFFECT OF FOREWARMING

To determine the effect on dispersibility, heat treatments were given flash-pasteurized whole milk of 3.2% butterfat, using conditions favoring the development of sulfhydryl groups *(2)*. The milk, after deaeration, was quickly heated to 190° F. in a Votator[2] and held at that temperature for approximately 2, 5,

[2] Mention of companies and commercial products anywhere in this paper does not imply that they are endorsed or recommended by the Department of Agriculture over others of a similar nature not mentioned.

and 8.5 min., respectively, then quickly cooled to below 100° F. These three batches, and a control given no heat treatment beyond that of flash pasteurization, were foam-dried by the published procedure *(3)*.

These were nitrogen-packed in cans with an oxygen content below 1.5% and stored at 100° F. for 1 mo. Dispersibilities were determined in 38° F. water with 60- and 100-sec. stirring. The results are shown (Table 2).

TABLE 2

Effect of forewarming of whole milk on its dispersibility after foam-drying[a]

Heat treatment		Dispersibility (%)[b]					
		60 sec. stirring			100 sec. stirring		
Time	Temp.	Fresh	Stored[c]	Loss	Fresh	Stored[c]	Loss
(min.)	*(°F.)*			*(%)*			*(%)*
0	.....	95.1	92.4	2.7	97.1	94.0	3.1
2.1	191	95.7	85.0	10.7	98.3	87.8	10.5
5.1	190	92.6	74.2	18.4	96.5	78.7	17.8
8.6	189	93.0	75.7	17.3	96.5	78.3	18.2

[a] All products contained 26% fat (MFB).
[b] Averages of data from four pilot-plant runs at each set of conditions.
[c] One month at 100° F.

It is apparent that the heat treatments had little effect on the dispersibility before storage. However, changes were induced which were responsible for a more rapid loss in dispersibility during storage than occurred in the product which had no heat treatment. The loss also increased with time of holding the heated milk. The losses in dispersibility were accompanied by a slight increase in insolubles suggesting that, as previously observed in the case of the unheated product stored at 100° F. (see Figure 2), loss in dispersibility was a consequence of protein insolubilization. It is important to avoid damage to the proteins if a dry *whole* milk of good dispersibility in ice water is to be produced. Damage may occur either during processing or on high-temperature storage of the dry product.

The importance of forewarming in improving the flavor stability of spray-dried whole milk is well known. The need of forewarming to protect against atmospheric oxidation during processing of foam-dried whole milk is unnecessary, because dehydration is carried out under high vacuum and low temperature, in contrast to the high-temperature oxidative conditions typically used in spray-drying. The latter conditions, coupled with the tenacious retention of entrapped oxygen within the spray-dried particle, create a different storage problem than is encountered with the foam-dried material. Moreover, preliminary tests using the forewarming conditions of Table 2 showed no improvement in the flavor stability of gas-packed, foam-dried material. The effects of forewarming and other factors on the flavor stability of the foam-dried product are currently under investigation.

The elimination of heat treatment (typically 180° F. for 30 min.) can pose a problem in controlling bacterial growth during operations. This can be over-

come by the sound practice of using milk of low bacterial count and by processing rapidly with only short-time holdup at points where temperature favors bacterial growth. For example, in the foam-drying process it was proposed to concentrate the milk at 90° F. *(3)*. If this were done in a conventional recirculation type evaporator, the relatively long time required to concentrate the milk would favor the growth of bacteria. Thus, this operation could better be carried out in a single-pass evaporator, wherein concentration is accomplished in a matter of seconds. This has the added advantage of permitting higher evaporation temperatures without protein denaturation. Evaporators of this type are characterized by the mechanical agitation of the heat-sensitive film on the heated surface. They are manufactured domestically by at least two companies. Another single-pass evaporator of possible use here is one developed at the Eastern Utilization Research and Development Division for fruit juices *(1)*.

Thus, the foam-drying process would be carried out somewhat as shown in Figure 3.

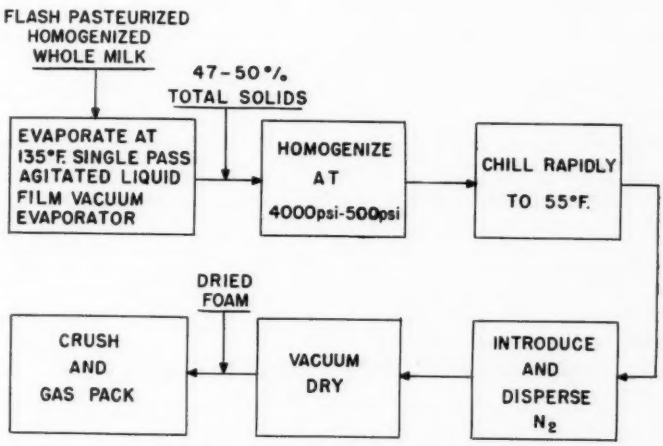

FIG. 3. Process for preparing foam-dried whole milk.

We have observed in the course of some 300 pilot-plant runs, drying whole milk, that when on rare occasions the solubility index was above about 0.1 cc., the otherwise excellent dispersibility of the dry whole milk was impaired. In contrast, it is well known that spray-dried *nonfat* milks of excellent dispersibility may contain 0.4 cc. insolubles or more. Similarly, in foam-dried nonfat milk, a higher level of insolubles can be tolerated without impairment of dispersibility than with dry whole milk. Therefore, the high-temperature processes commercially feasible with nonfat milk are at a disadvantage with whole milk.

Foam-drying of other fat-containing milks. Adaptation of the foam-drying process to fat-containing milks other than whole milk was shown to enhance the dispersibility of these products. They include a chocolate-flavored whole milk

and infant foods such as lactose-enriched whole milk and an acid-modified whole milk. Table 3 gives pertinent drying data on these products. Evaporation temperature and the temperature and pressures of homogenization were, in all cases, the same as in the standard process for whole milk.

TABLE 3

Conditions used in foam-drying fat-containing milk products

Material whole milk	Total solids in concentrate	Drying			Product moisture
		Time	Max. temp.	Volume increase	
	(%)	*(min.)*	*(°F.)*	*(fold)*	*(%)*
Chocolate-flavored whole milk	50.70	207	107	32	1.33
Lactose-enriched whole milk	55.38	161	106	24	1.80
Acid-modified whole milk	40.85	118	114	10	1.03

1. *Chocolate-flavored whole milk.* A commercially available chocolate-flavoring powder composed of Dutch-processed cocoa, sugar, salt, an emulsifier, and flavorings was added to whole milk of 4.1% butterfat, 12.9% total solids, in the ratio of 1 to 25.8 lb. The mixture was evaporated, homogenized, gassed, chilled, foam-dried, and crushed through a 20-mesh screen in the usual manner *(3)*.

2. *Lactose-enriched whole milk.* Lactose monohydrate was dissolved in whole milk of 4.1% fat, 12.9% total solids, in the ratio of 1 to 19 lb. This resulted in a dry product similar in composition to commercial products of this type. The formulation was then evaporated, gassed, chilled, foam-dried, and crushed through a 20-mesh screen in the usual manner *(3)*, except that evaporation was carried beyond 50% solids. This was possible because at 50% solids the concentrate showed no tendency to gel and was more fluid than whole milk at the same solids content.

3. *Acid-modified whole milk.* The dry product was prepared from the following recipe:

	(lb.)
Whole milk (4.1% fat, 13.0% solids)	41.24
Dextrins-maltose	1.21
Sucrose	1.21
Corn starch	0.83
Lactic Acid (85% soln.)	0.24

The resulting dried product was similar in composition to commercial products of this type. The milk was warmed to 90° F. and the ingredients added with stirring. Curdling occurred just before all of the acid had been added. In spite of this, the formula was processed in the usual manner *(3)*, except that evaporation was carried only to about 41% solids, due to the high viscosity of the concentrate at that point. A subsequent experiment showed that a solids

content of about 38% yields a foam of more desirable physical form than was obtained in the present instance.

Dispersibility of the dry materials. Figure 4 contains dispersibility rate curves for the three foam-dried products and, in the cases of the infant foods,

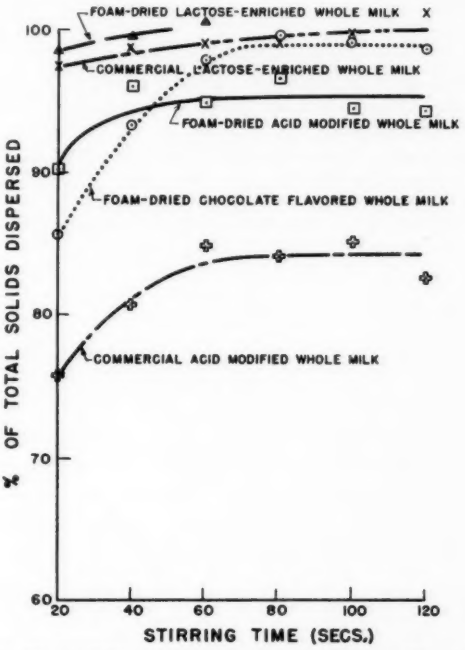

FIG. 4. Dispersibility rates of dry fat-containing milk products.

for commercially available products of similar compositions. These curves were obtained by use of the dispersibility rate test as previously published *(3)*, with the following exceptions:

1. The weight of the dry chocolate-flavored whole milk was increased from 15 to 17.4 g. per 90 cc. of water. This was done so that its dispersibility curve would be based on the solids content of a typical chocolate-flavored whole milk.
2. The infant foods were dispersed in water at 100° F. instead of the usual 38° F., because in household practice water at or above the former temperature would normally be used for reconstituting products of these types.

The curves show that foam-drying can be advantageously applied to these materials. The curves show that a dry chocolate-flavored whole milk of excellent dispersibility may be prepared by foam-drying. The results suggest the application of foam-drying to whole milk flavored with fruit juice concentrates, es-

sences, honey, or other natural flavors. The authors are currently investigating this.

ACKNOWLEDGMENT

The authors express their appreciation to R. E. Townend of the Eastern Utilization Research and Development Division for his ultracentrifugal studies and to R. W. Whittenberger of the Eastern Utilization Research and Development Division for his microscopic observations.

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RAPID DETERMINATION OF MILK SALTS AND IONS. I. DETERMINATION OF SODIUM, POTASSIUM, MAGNESIUM, AND CALCIUM BY FLAME SPECTROPHOTOMETRY

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Research Laboratories of Nestlé Products, Vevey, Switzerland

SUMMARY

A rapid determination of milk cations is presented. A trained technician can make four to six complete analyses a day for sodium, potassium, calcium, and magnesium. The cation exchange resin Dowex 50 is used to eliminate, quantitatively, the total amount of cations from a milk serum prepared according to the procedure of Jenness *(2)* and Murthy and Whitney *(4)*. Then, the cations are quantitatively eluted with 3.5 *N* HCl and analyzed by flame spectrophotometry. The accuracy of the method is due to the fact that the analyzed solution does not contain other substances than the cations to be analyzed—hydrochloric acid and ammonia. The interference of phosphoric acid, especially, and of organic materials, is avoided.

Many attempts have been made to determine accurately and rapidly the mineral components of milk. However, it seemed difficult to apply one single method to the analysis of the four major cations in milk, sodium, potassium, magnesium, and calcium. Sutton and Almy *(7)* have used successfully the ion exchange chromatography technique to separate these four cations, with subsequent titration of the pure chlorides. This procedure includes ashing of the milk samples, chromatography, evaporation of excess HCl, and titration of chlorine; consequently, it is long and tedious. Keirs and Speck *(3)* described the widely used flame spectrophotometric technique of milk ash solutions, and they give a good over-all view of the problem. The paper of Riethmüller *(5)* is referred to for the fundamentals of emission spectrophotometry of the elements and for the most suitable equipment and techniques.

To avoid the time-consuming ashing of milk samples, the technique of Jenness *(2)*, modified by Murthy and Whitney *(4)*, was applied to get a casein-free milk serum containing all cations present in the milk. Jenness *(2)* was interested only in Mg and Ca analysis by the titration method of Schwarzenbach *et al. (6)*. Murthy and Whitney *(4)* used their milk serum for direct flame spectrophotometric analysis of Na, K, and Ca. Keirs and Speck *(3)* stated that the phosphoric ion interferes seriously with the Ca flame analysis. To overcome this difficulty, Murthy and Whitney *(4)* eliminated an important part of the interfering anions with anion exchange resins. The sample ready for analysis still contained large amounts of organic material, making it impossible to determine magnesium accurately.

The procedure described in this paper made it possible to analyze four to six milk samples a day with accuracy, and the same sample is used to analyze the four cations, Na, K, Mg, and Ca.

Received for publication December 18, 1957.

METHOD

Instrument. For preliminary trials, a Beckman B spectrophotometer with flame attachment has been used. The present study refers to data obtained with a Beckman DU spectrophotometer with flame attachment and photomultiplier. The instrument was operated with a hydrogen-oxygen burner as indicated in Table 1. The instrument allows accurate measurements with a maximum deviation of single determinations of $\pm 2\%$.

TABLE 1

Suitable conditions for flame spectrophotometric analysis of Na, K, Ca, and Mg

Analyzed element	Suitable concentration range	Wave length	Photo tube	Sensitivity		Slit width	Gas pressure	
				DU	Photomultiplier		H_2	O_2
	(p.p.m.)	*(mμ)*				*(mm.)*	*(p.s.i.)*	
Potassium	0 to 60	767	Red	0	..	0.01	2	10
Sodium	0 to 10	588	Blue	0	2	0.01	2	10
Calcium	0 to 60	554	Blue	0	3	0.04	2	10
Magnesium	10 to 60	371	Blue	0	3	0.20	4	15

Preparation of the milk serum. The procedure of Murthy and Whitney *(4)* with minor variations was used. To 60 ml. of fresh milk or 7.5 g. of dried whole milk, water is added to a volume of approximately 100 ml. The casein is precipitated by adding five ml. of 1 *N* HCl and the solution is allowed to stand for 10 min. The sample is adjusted with 1 *N* NH_4OH to pH 4.0–4.1 and the volume made up to 150 ml. The solution is filtered through an analytical-grade filter paper or centrifuged. A 100-ml. sample, equivalent to 40 ml. of fresh milk or to 5 g. of dried whole milk, is collected for analysis of the cations. The casein-free filtrate can be obtained, in variation of this procedure, by adding to the diluted milk 2 ml. of glacial acetic acid instead of 5 ml. of 1 *N* HCl. In this case, subsequent heating at 80° C. for 10 min. is necessary, but adjusting the pH with ammonia can be omitted. In this study, the HCl precipitation procedure was used.

Preparation of the pure chloride solution of cations. The 100-ml. sample of casein-free acid serum was passed quantitatively through a column of 10 ml. of Dowex 50 cation exchange resin in the hydrogen form. We used uniform particle-size of the resin passing through 30-mesh screen (wet) and retained on a 40-mesh screen. The columns were 30 cm. long, with an inner diameter of 0.9 cm., and were closed with a thin-tipped stopcock, and the resin was retained by a small plug of glass wool just above the stopcock. The sample was passed through the column at the rate of 4 ml. per minute, then the resin was washed with three portions of 10 ml. of distilled water until the pH of the effluent was neutral.

During the adsorption cycle of the cations, the absence of cations in the effluent solution is checked by the flame test, with a drop of the solution taken from the bottom of the column with a platinum wire. Since the cation-binding capacity

of the Dowex 50 column should be about double the cations to be adsorbed, no cations should be detectable during adsorption, including the washing with water.

By this procedure, all anions and all organic substances are eliminated from the sample to be analyzed. The cations are eluted with 75 ml. of 3.5 *N* HCl at a rate of 2 ml. per minute. The first 3 ml. of the effluent, consisting of water only, are discarded. The following 50 ml. of acid solution, which contains the cations (Figure 1), are collected in a 50-ml. volumetric flask. The end of the elution is verified by the flame test, with a platinum wire. Traces of calcium still may be present after 50 ml. of acid has passed through the column, but are without significance for the quantitative analysis of Ca.

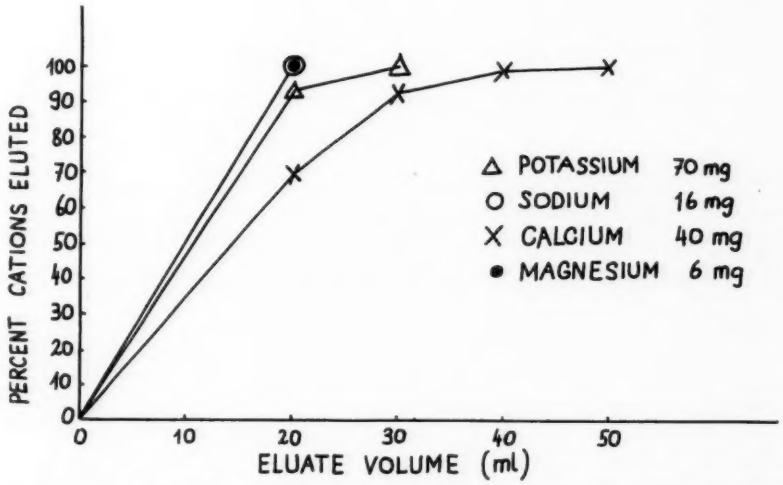

FIG. 1. Elution with 3.5 *N* HCl of potassium, sodium, calcium and magnesium, adsorbed on 10 ml. of Dowex 50 cation exchange resin.

The columns are then ready for a new adsorption cycle, after the complete elution of the cations with 3.5 *N* HCl and after rinsing the column with distilled water to eliminate the concentrated acid.

This procedure is to some extent similar to the method described by Denson *(1)*, who eliminated the phosphoric acid from solutions of dissolved ashes derived from biological material by adsorbing the cations on an Amberlite IR-105 cation exchange resin. (Denson reported that he also had trouble with the elution of calcium.)

Flame photometric determination of Na, K, Mg, and Ca. The hydrochloric acid solutions of the cations are thoroughly mixed and diluted to appropriate concentrations. For Na and K analysis, a dilution of 1:100 in 0.5 *N* HCl is suitable, yielding concentrations of from 1 to 10 p.p.m. of Na and of from 3 to 60 p.p.m. of K (Figures 2 and 3). For Ca analysis, a dilution of 1:25 in 0.5 *N*

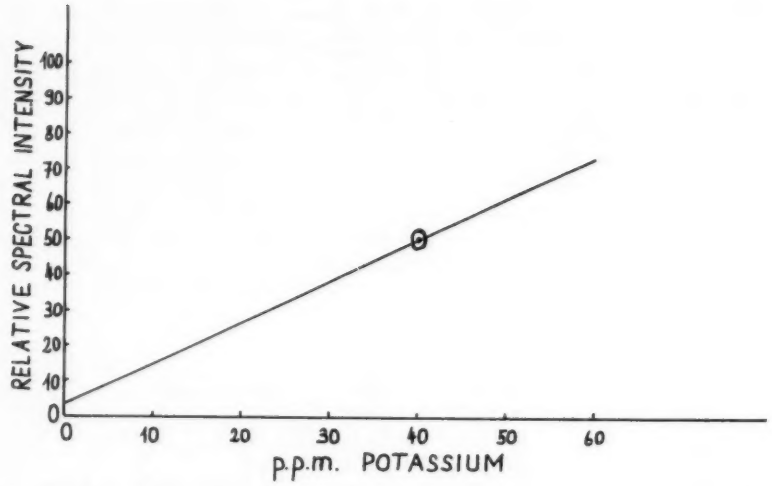

FIG. 2. Calibration curve for potassium. The curve is obtained by measuring the potassium emission (Table 1) of 12 concentrations of K ranging from 5 to 60 p.p.m., added to a standard solution containing 3.5 p.p.m. of Na, 8 p.p.m. of Ca, 1.2 p.p.m. of Mg, and 14 p.p.m. of NH_4OH in 0.5 *N* HCl.

⊙ = reference point arbitrarily chosen to check the photometer before running an analysis.

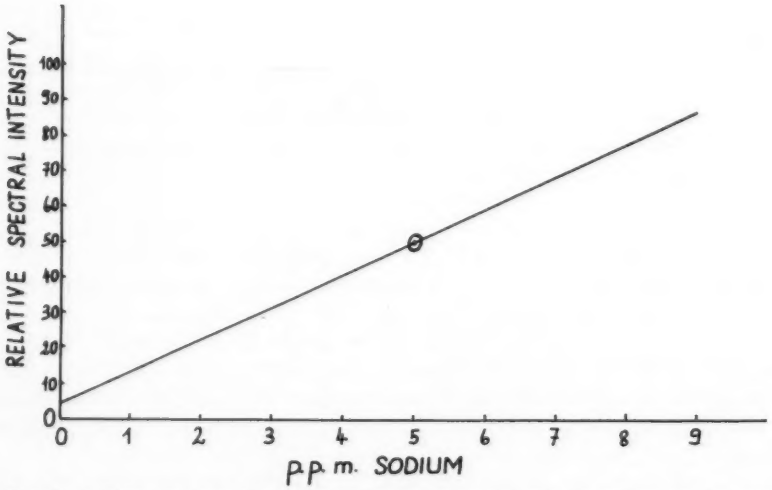

FIG. 3. Calibration curve for sodium. The curve is obtained by measuring the sodium emission (Table 1) of ten concentrations of Na ranging from 1 to 10 p.p.m., added to a standard solution containing 14 p.p.m. K, 8 p.p.m. Ca, 1.2 p.p.m. Mg, and 14 p.p.m. NH_4OH in 0.5 *N* HCl.

⊙ = reference point arbitrarily chosen to check the photometer before running an analysis.

HCl is the most suitable dilution, yielding concentrations of from 5 to 50 p.p.m. (Figure 4). For Mg analysis, a dilution of 1 : 5 with water is made up, yielding concentrations of from 10 to 60 p.p.m. of Mg in a 0.7 *N* hydrochloric acid solution (Figure 5). Before making up the dilution for Mg, the three cations Na, K, and Ca are analyzed. Then, with these known concentrations, a dilution of the sample for Mg analysis is calculated, to get close to the concentration ranges of cations of the standard solution chosen for the Mg calibration curve. It is most important that the total concentration of Na plus K plus Ca correspond as closely as possible to the concentration of 510 p.p.m. of these three cations, as used to establish the calibration curve for Mg (Figure 5). Ammonia has practically no influence in a wide concentration range.

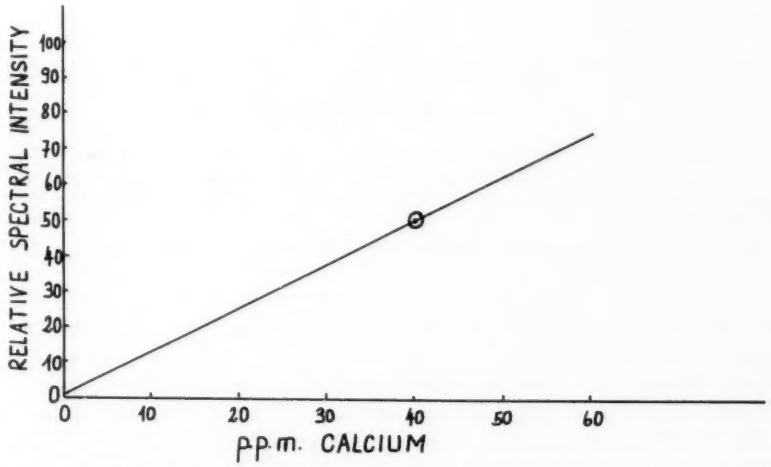

FIG. 4. Calibration curve for calcium. The curve is obtained by measuring the calcium emission (Table 1) of 12 concentrations of Ca ranging from 5 to 60 p.p.m., added to a standard solution containing 56 p.p.m. K, 14 p.p.m. Na, 4.8 p.p.m. Mg, and 56 p.p.m. NH_4OH in 0.5 *N* HCl.

⊙ = reference point arbitrarily chosen to check the photometer before running an analysis.

The diluted pure chloride solutions are analyzed with the Beckman DU spectrophotometer under conditions as pointed out (Table 1). Results of the analyses, read on the transmission dial, are compared to a calibration curve.

Calibration curves (see Figures 2, 3, 4, and 5). It is important to establish the calibration curves carefully. Every constituent in the sample to be analyzed should be present in the standard solutions used for the calibration, because every substance present interferes with the specific emission of a certain concentration of an element in the flame. For the same reason, no other substance is allowed to be present in the standard solution. Therefore, all standard solutions contain HCl, K, Na, Ca, Mg, and ammonia, in concentrations which are expected to occur in the sample to be analyzed.

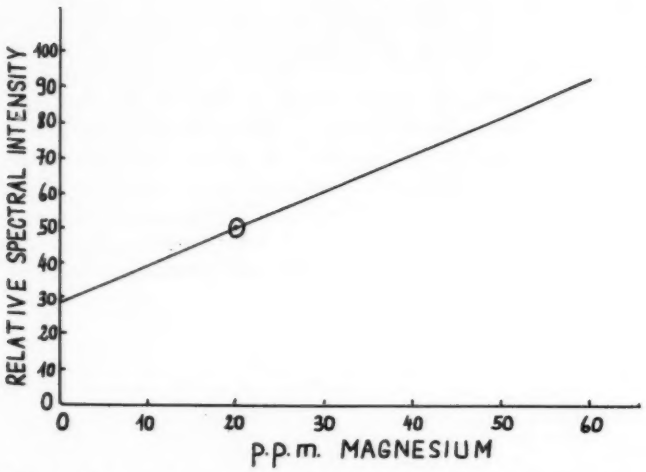

FIG. 5. Calibration curve for magnesium. The curve is obtained by measuring the magnesium emission (Table 1) of 12 concentrations of Mg ranging from 5 to 60 p.p.m., added to a standard solution containing 280 p.p.m. K, 70 p.p.m. Na, 160 p.p.m. Ca, and 280 p.p.m. NH_4OH in 0.7 *N* HCl.

⊙ = reference point arbitrarily chosen to check the photometer before running an analysis.

The calibration curves shown (Figures 2, 3, 4, and 5) are typical for analyses of milk, and they can be used only for those cations and in solutions of this standard composition.

Once the calibration curves are established, one standard solution is chosen to balance the DU spectrophotometer each time before the unknown samples are analyzed. With this standard solution, the transmission dial is set to the reference point on the calibration curve and the photometer balanced; a slight adaptation of the slit width may be necessary for accurate reproduction of standard conditions.

RESULTS AND DISCUSSION

Reproducibility of the flame photometric analysis and recovery of cations after adsorption on the Dowex 50 ion exchange resin. The trials on reproducibility and recovery experiments are shown (Figure 1 and Table 2). In Figure 1 it can be seen that known amounts of cations as chlorides, dissolved in a 100-ml. sample, had been adsorbed on Dowex 50 (see also Table 2, Experiment 1). During the elution with 3.5 *N* HCl, four fractions of 20, 10, 10, and 10 ml. had been taken and each of them analyzed for Na, Mg, K, and Ca. Under these conditions, 20 ml. of HCl are sufficient for a 100% recovery of Mg and Na; the recovery of K was complete after elution of 30 ml. of HCl, and Ca needed 50 ml. for total recovery.

Table 2, Experiment 2, shows that a salt mixture added to skimmilk is recovered quantitatively after going through the complete analytical procedure; in Experiment 3, the maximum deviations of Na, K, and Ca on the mean values

TABLE 2

Reproducibility and recovery experiments of the flame photometric analysis of cations

Experiment No.	Sample analyzed		Cations adsorbed on Dowex 50 calculated	Cations eluted from Dowex 50 found	Recovery	Maximum deviation
			(mg.)		(%)	
1	Salt mixture	K	70.0	71.0	101	+ 1
		Na	16.0	15.6	97	− 3
		Ca	40.0	40.8	102	+ 2
		Mg	6.0	5.9	98	− 2
2	40 ml. skimmilk[a]	K	(?)	65.0		
		Na	(?)	14.0		
		Ca	(?)	50.0		
		Mg	(?)	4.2		
	20 ml. skimmilk with added salt mixture[a]	K	64.5	64.0	99	− 1
		Na	14.0	13.5	96	− 4
		Ca	55.0	55.0	100	0
		Mg	4.1	3.8	93	− 7
3	Means of five different samples of 100 ml. of skimmilk	K	(?)	168		± 5.8
		Na	(?)	35		± 5.5
		Ca	(?)	119		± 7.5
		Mg	(?)	11		±12.7
4	10 g. whole milk powder *(c)*, means of three complete analyses[b]	K	(?)	122.0		± 0
		Na	(?)	24.7		± 3
		Ca	(?)	95.3		± 1
		Mg	(?)	8.8		± 2
	10 g. whole milk powder *(c)*, ashed before adsorption on Dowex 50[b]	K	122.0	122.0		0
		Na	24.7	22.0		−11
		Ca	95.3	94.0		− 1.4
		Mg	8.8	8.4		− 0.5

[a] Identical samples of skimmilk.
[b] Identical samples of whole milk powder.
[c] Nestlé whole dry milk powder (Nido).

of skimmilk analysis are larger than the error on the recovery experiments. This means that the method is suitable for detecting variations in the cation content of milk. Experiment 4 shows an excellent reproducibility of the two different preparations of the milk sample, after casein precipitation with acid as described by Jenness *(2)* and Murthy and Whitney *(4)*, and after ashing of the milk powder. Ashing of milk at temperatures above 400° C. often produces losses of Na and Cl, as observed in our Laboratory (unpublished data). This may explain the lower figure found for Na in milk ash determination.

Interference of cations in the flame spectrophotometric analysis. All constituents of a solution to be analyzed by flame spectrophotometry interfere more or less with the determination of cations. For this reason, the calibration curves should contain all constituents of the sample to be analyzed and in the same proportions. Therefore, it is important that the sample to be analyzed be purified from all undesired ingredients, as far as is possible; care should be taken, especially, not to introduce new ones. The cation adsorption on Dowex 50 with

subsequent elution with HCl seems to be an especially important step, because of the complete elimination of organic substances and the complete elimination of all anions. An accurate Ca and Mg analysis is impossible if the phosphoric acid is not eliminated quantitatively.

By this process of cation adsorption on Dowex 50, the reciprocal interferences of the cations themselves need to be considered, as well as the minor influences of HCl and NH_4OH. The concentrations of HCl and NH_4OH are held constant; therefore, they do not interfere. The ammonia, especially, has little influence.

The interference of cations is of two kinds: *(a)* Depression of emission by the presence of high concentration of other cations: *(b)* increase of the emission by other cations.

The depression of emission does not appear under the conditions described in this paper; the total concentration of cations does not exceed 1,000 p.p.m. (see Figure 5, calibration curve for Mg).

The increase of emission of one cation by other cations always is to be expected. For the Na, K, and Ca determination, those interferences would be of minor importance, because of the low total concentration of the cations and the high emission activities of these ions. For Mg, the interferences of Na, K, and Ca are important, because of the weak emission intensity of Mg and the relatively small proportion of Mg present in milk minerals, the total concentration of Ca, K, and Na being about 30 times as high as that of Mg. For accurate Mg analysis, therefore, it is necessary to analyze Na, K, and Ca first, then to calculate the concentration range of the sample and dilute to a concentration of Na plus K plus Ca, so as to obtain a concentration range as close as possible to the concentration used to establish the calibration curve. It has to be considered that the HCl content of the final dilution must be 0.7 *N* (see Figure 5).

ACKNOWLEDGMENT

The author gratefully acknowledges the help of Dr. E. Bovet of the Federal Experimental Station of Agriculture at Lausanne, Switzerland, while starting work on flame spectrophotometric determination of cations.

The analytical work was done by F. Moser, Laboratory Assistant, who contributed much.

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COMPARISON OF STANDARD METHODS OF MEASURING HOLDING TIME IN H.T.S.T. PASTEURIZERS

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SUMMARY

Three tests for the determination of holding time were compared; namely, the nickel chloride–dimethylglyoxime chemical method, the sodium nitrite–Griess-Ilosvay chemical method, and the salt conductivity method. Tests were carried out, under field conditions, on five commercial plate-type, high-temperature, short-time pasteurizers, the capacities of which ranged from 3,935 to 16,525 lb. per hour. The size of holding tubes on these units was from 1½- to 2½-in. nominal size sanitary piping, whereas one of the units had a plate-type holder section.

The methods were compared under conditions as nearly identical as circumstances would permit. Results of tests, carried out in accordance with these methods, were compared on the basis of the same flow rate, with the same type of injection being used. The point of injection of the solution used as a tracer was the same in all tests in relation to the flow of liquid entering the holding section.

The results indicate that the sodium nitrite test was more sensitive as a method of determining holding time than either the nickel chloride test or the salt conductivity test. This was more apparent in high-capacity units, using larger-sized holding tubes, and in lower-capacity units with a low holding-tube efficiency. The nickel chloride test and the salt conductivity test were shown to be of comparable sensitivity on low- and medium-capacity units, but on higher-capacity units the nickel chloride test appeared more sensitive than the salt conductivity test.

The universal adoption of H.T.S.T. pasteurization of milk, with a holding time of a matter of a few seconds, necessitates a precise and accurate method of measuring the time of holding of the milk under this system. Various methods have been used or suggested for use in the determination of holding time in H.T.S.T. pasteurizers, each having its relative advantages and disadvantages. The object of any method is to determine the minimum time in which a bacterial cell will travel through the holding tube. Recent years have seen the development of more accurate methods. This study is a comparison of two methods of determining holding time, one of which is considered to be the most accurate method available, and the other, one of the most widely used methods. The former method may be called a chemical method as it involves the injection of a chemical substance at the entrance to the holding tube which is detected at the outlet of the holding tube by a reagent which gives a colour reaction. The latter method is the electrolytic or salt conductivity method which has been standardized in the United States for the measurement of holding time in H.T.S.T. pasteurizers.

REVIEW OF THE LITERATURE

The fact that certain chemicals can be detected by a simple colour reaction forms the basis of a test for holding time devised by Dummett and Mongar *(3)*. This method depends on the injection of a saturated solution of nickel chloride and employs the organic reagent dimethylglyoxime as a detector, which gives

Received for publication January 9, 1958.

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a yellow colour or pink precipitate according to the concentration. In carrying out the test, 5 cc. of the saturated solution of nickel chloride are injected at the inlet to the holding tube by a spring-loaded syringe. A sampling valve is fitted at the outlet of the holding tube. Samples are taken at 1-sec. intervals into the sampling tubes containing 1 cc. of dimethylglyoxime solution. The presence of nickel chloride in any filled tube will be shown by the colour reaction with the reagent. The holding time in seconds thus is automatically recorded by the number of clear tubes, plus the time in seconds from injection to start of sampling. The sensitivity of this method is claimed to correspond to a dilution of 1 part of nickel chloride in 100,000 parts of water.

A variation of the Dummett and Mongar test was suggested by Botham *(1)*, who used sodium nitrite as the injected solution and the Griess-Ilosvay reagent as the detector. Sodium nitrite was chosen by Botham as a chemical which could be used to test the holding time on milk. This test was shown in laboratory experiments, with milk warmed to pasteurizing temperature, to give a recognizable pink colour in the presence of as little as one part of added sodium nitrite in two million parts of milk. In a series of tests run on the latest design of holding tube, using water and milk successively, it was shown that, whereas the nickel chloride test gave 17 sec., the sodium nitrite test gave a holding time of 16½ sec. Botham claims that the holding time for a particular piece of apparatus in turbulent flow conditions is given with sufficient accuracy both by nickel chloride and dimethylglyoxime on water, or by sodium nitrite and the Griess-Ilosvay reagent on milk. Burton and Thiel *(2)* describe the nitrite test as the most sensitive test yet available for determining holding time in H.T.S.T. pasteurizers.

Electrolytic methods of determining holding time are adapted from methods of measuring water-flow in pipes. The principles of the electrolytic flow measurement in application to H.T.S.T. pasteurizing units are given by Roger *(7)*. A saturated solution of sodium chloride was injected at the inlet to the holding tube where a pair of electrodes, joined in circuit with dry cells, and a 25 Ma. meter were situated. Another pair of electrodes connected to a similar circuit, in which a 5-Ma. meter was used, was situated at the outlet of the holding tube. In operation, one person injects the salt solution and another starts the stop watch when the needle of the ammeter in the circuit at the inlet deflects owing to the increased conductivity caused by the salt charge passing the electrode; then stops the watch when the ammeter needle at the outlet shows a deflection. A detailed description of the electrolytic method is given by Fay and Fraser *(4)*, who compared the manual system of timing with timing by means of an automatic electric timer. Over a series of 11 readings, they found a maximum variation of 1 sec. with manual timing, and 0.6 sec. with automatic timing. Weber *(9)* observed that the salt test is a practical means of determining the minimum holding time when operating with water. It has the disadvantage that it is quite complicated, and can not be used directly on milk or milk products. Robinson and Moss *(6)* note that the detection of a salt charge in the holding tube was somewhat slower by an automatic timer than by a milliammeter. The difference

in time between the two instruments varied from 0.9 to 3.50 sec., and the difference increased with decreasing velocity of flow. A study of possible variations which might be introduced into the salt test was made by Jordan *(5)*. He found that for an average velocity of water at 160° F. of 1.268 ft. per second in 2½-in. sanitary pipe, all injected volumes of saturated sodium chloride solution from 15 to 150 cc. gave holding times which agreed within about 0.1 sec. A decrease in the injected volume below 15 cc. apparently resulted in increasing the holding time. Jordan also observed that the direction in which the salt solution was introduced into the tube affected the apparent holding time. A difference in apparent holding time of about 0.7 sec. was found between injections with, and injections against, the stream. A spray-type nozzle and a straight tube injection across the flow were found to give comparable results. When compared under identical conditions, manual (or stop watch) timing and automatic timing were found to give holding times which agreed closely. From the results of a series of experiments showing that the organism, *Escherichia coli*, and the sodium chloride are transported at the same rate by water at 80° F., Jordan concludes that, if satisfactorily sensitive means are used for detection, salt can be used for the estimation of the holding time of bacteria.

The necessity of having a standardized test for holding time was recognized in the United States in 1950, when the 3A Standard method for determining the holding time in H.T.S.T. pasteurizers by the salt conductivity test was formulated. This standard gives approved apparatus and materials to be used in the test and outlines the method to be followed, step by step. Formulae are also given for computing the holding time for milk from that obtained when the unit is operated on water.

EXPERIMENTAL PROCEDURE

This comparative study was carried out on selected plate-type pasteurizing units. The holding time was determined on each unit tested by means of the nickel chloride–dimethylglyoxime test of Dummet and Mongar *(3)*, the sodium nitrite–Griess-Ilosvay test of Botham *(1)*, and the 3A Standard salt conductivity test *(8)*.

The equipment used in the chemical tests, that is, the nickel chloride and sodium nitrite tests, was manufactured as a standard test-kit by the A.P.V Company Ltd., Sussex, England. Minor alterations were made on the injection and sampling devices supplied, to ensure that the point of injection or sampling came to the center of the flow stream through any size of sanitary connections to which the devices were fitted. Spray-type injection nozzles were used in all tests, because this method of injection is considered most accurate under varying circumstances and is also the method adopted in the 3A Standard salt conductivity test. Holding time was determined from the results of six chemical tests on each unit tested. In three of these tests, sampling was begun 10 sec. after injection of the tracer and in the remaining three, 10½ sec. after injection. This allowed the holding time to be expressed to the nearest 0.5 sec. After each test the unit was run for approximately 5 min., to ensure that the injected solution was cleared from the system.

Preliminary tests were carried out to determine the sensitivity of the reagents used in the chemical tests. These confirmed the claims made in respect to the tests; namely, that the Griess-Ilosvay reagent is capable of detecting as little as one part of sodium nitrite in 10 million parts of water, and that dimethylglyoxime is capable of detecting as little as one part of nickel chloride in 100,000 parts of water, under practical operating conditions.

The equipment and method used in the salt conductivity test conformed to the standards specified in the 3A Standards for this test. The electrodes and automatic Solu-Bridge Flow Timer employed in the tests were manufactured by Industrial Instruments Inc., New Jersey, U.S.A. The automatic Solu-Bridge Flow Timer responded to a concentration as low as 4 p.p.m. of salt in water. A response to at least 10 p.p.m. of salt in water is specified for the conductivity instrument used in the salt test by the 3A Standard method. The holding time on each unit tested by the salt conductivity test was taken as the average of ten successive tests, with a variation between the minimum and maximum values not exceeding 0.5 sec.

Holding-time determinations, by the methods outlined, were made on commercial plate-type, high-temperature, short-time pasteurizers, selected to give a wide range of capacities and size of holding tube. Five units were included in the study, having capacities of 3,935 to 16,525 lb. per hour. The units had tubular holding sections with one exception; this unit had a box plate-type holding section incorporated in the press.

During test runs, the units were operated on water at approximately milk pasteurization temperature, with a constant head of water maintained in the pump feed-tank during operation. The holding time on each unit was first determined by the chemical method, the unit closed down and the injection nozzle and sampling valve replaced by the salt conductivity test electrodes. On restarting, the holding time was determined by the salt conductivity test. Holding-time determinations were done only after the unit had attained equilibrium temperature and flow conditions.

The flow rate of the unit was checked regularly during the period of the test run. The discharge from the sampling valve during the chemical test was taken into account in computing the flow rate of the unit during that test. However, a discrepancy was noted in the flow rates of the units computed during the periods when the two tests were being carried out, even though the units were being operated under identical conditions. It was concluded, therefore, that the discrepancy, an increase in the flow rate during tests by the chemical method, was caused by the release of pressure within the system, due to the discharge through the sampling valve used in the chemical method. This had the effect of reducing the discharge head of the unit, causing the discharge of the pump to increase. This effect was greatest when a Waukesha positive displacement pump was used; whereas, the discharge was much more constant when a homogenizer was used.

In order that the holding times obtained by the different methods could be compared on the basis of the same flow rate, it was decided to adjust the holding

times given by the salt test, to the higher flow rate measured under conditions of testing by the chemical test. The validity of carrying out this adjustment was established by a series of tests carried out on a plate-type holding section. These tests showed that the holding time given by the salt conductivity test could be adjusted by direct calculation to the higher flow rate measured during the chemical test, within a limited range of about 100 lb. per hour. This was the order of the discrepancy in flow rate taken during the two tests. Thus, the holding time given by the salt conductivity test on the units included in this study has been adjusted in this manner to give a test comparable with the holding time given by the chemical method. In the presentation of results, this adjusted salt conductivity test has been given to the nearest lower 0.5 sec. This was done to give a truer comparison of the tests, because the chemical test results also are expressed in this way due to the method of sampling.

The efficiency of the holding tube of each unit tested was calculated, using the following formulae:

$$\bar{v} = \frac{w}{\rho\, a\, t} \tag{1}$$

where $\bar{v}$ = average velocity, ft/sec
w = weight of water collected, lb.
ρ = density of water at measured temperature, lb/ft^3
a = area of pipe, ft.^{2}
t = time for collecting water, sec.

$$v_{H.T.} = \frac{L}{H.T.} \tag{2}$$

where $v_{H.T.}$ = velocity of fastest measured particle, ft/sec
L = length of tube between injection and detection point, ft.
H.T. = measured holding time, sec.

$$\text{Holding-tube efficiency} = \frac{\bar{v}}{v_{H.T.}} \tag{3}$$

All holding-tube efficiencies were calculated on the basis of the holding time measured by the Griess-Ilosvay test and the flow rate measured during that test. When obtaining the length of holding tubes, the equivalent length of straight piping for 90°-sweep ells in the tube was taken from measurements given by the New York State Department of Health.

RESULTS AND DISCUSSION

The following results are given for each individual pasteurizing unit, together with any relevant details of the unit:

Unit No. 1. The holding tube of this unit was 30.82 ft. long, in 1½-in. sanitary pipe with C.I.P. fittings. The calculated efficiency of the tube was 87%. The milk pump was a Creamery Package homogenizer, driven by a 10-h.p. electric motor, having a maximum operating pressure of 2,000 p.s.i.g. During tests, the homogenizer was operated with no pressure on the homogenizing valve. The unit was operated on water at 163° F. ± 0.5° at a flow rate of 3,935 lb. per hour.

The results of the holding time tests are given (Table 1). They indicate that, under these circumstances, the three tests have similar sensitivities.

TABLE 1
Summary of holding time tests

Unit	Flow rate	Holding tube efficiency	Holding time		
			Sodium nitrite	Nickel chloride	Salt conductivity
	(lb/hr)	*(%)*	*(sec.)*		
1	3,935	86	16	16	16
2	4,274	75	18	18.5	18.5
3	8,001	77	16	17	16.5
4	13,153	86	14	14.5	14.5
5	16,525	83	17.5	18	18.5

Unit No. 2. The holding tube of this unit was 15.10 ft. long, of three sizes of sanitary piping; namely 2, 2½, and 3 in. The calculated efficiency of the tube was 74.6%. The milk pump was a Waukesha positive displacement pump, with Reeves variable speed drive, driven by a 1.5-h.p. electric motor. The unit was operated on water at 165° F. ± 0.5° at a flow rate of 4,274 lb. per hour. The results are given (Table 1).

Under these conditions, the results indicate a higher sensitivity of the sodium nitrite test and a lower, but comparable, sensitivity of the nickel chloride and salt conductivity tests.

Unit No. 3. A plate-type pasteurizing unit in which the holding section was made up of eight, ¾-in. holder plates, which consisted of a central frame enclosed by two plates of design similar to those used in other sections of the unit. The calculated efficiency of the holder section was 77.2%. The milk pump was a Waukesha positive displacement pump with Reeves variable speed drive, driven by a 1.5-h.p. electric motor. The unit was operated on water at 163° F. ± 0.5°, at a flow rate of 8,001 lb. per hour. The results of holding-time tests on this unit are given (Table 1).

These results show the sodium nitrite test to have a higher sensitivity than the nickel chloride test and the salt conductivity test. The salt conductivity test is shown to give a holding time of 0.5 sec. less than the nickel chloride test. It is pointed out in this case, however, that the salt conductivity test after adjustment to flow rate was 16.83 sec. Under the system selected for presenting results, this became 16.5 sec. Thus, it is probable that the holding times measured by the salt conductivity test and the nickel chloride test were similar under these conditions.

Unit No. 4. The holding tube of this unit was 51.09 ft. long, in 2-in. sanitary piping with C.I.P. fittings. The calculated efficiency of the tube was 86%. The milk pump was a Cherry-Burrell homogenizer, driven by a 50-h.p. electric motor, with a maximum operating pressure of 2,500 p.s.i.g. During tests, the homogenizer was operated on water at 163° F. ± 0.5° at a flow rate of 13,153 lb. per hour, and with no pressure on the valve. Results are given in Table 1.

These results indicate that the sodium nitrite is more sensitive than the nickel chloride and salt conductivity tests, under conditions of high holding-tube efficiency, with larger flow rates. The controlling factors in this case ap-

peared to be the greater dilution of the injected tracer in the holding tube, and the greater sensitivity of the sodium nitrite–Griess-Ilosvay reaction, enabling earlier detection of the foremost particles of tracer passing through the tube.

Unit No. 5. The holding tube of this unit was 51.56 ft. long, in 2½-in. sanitary piping. The calculated efficiency of the tube was 83.3%. The milk pump was a Waukesha positive displacement pump, with Reeves variable speed drive, driven by a 2-h.p. electric motor. The unit was operated on water at 163.5° F. ± 0.5°, at a flow rate of 16,525 lb. per hour. Results are given (Table 1).

These results bear out the results found with Unit No. 4. In this case, with the larger size, 2½-in. holding tube, and a high holding-tube efficiency, the sodium nitrite test again is shown to exhibit a higher sensitivity of holding-time determination than either the nickel chloride test or the salt conductivity test. In addition, under these conditions, the nickel chloride test is shown to be more sensitive than the salt conductivity test.

CONCLUSIONS

The relative sensitivity of the tests studied is a function of the size of the unit and the size and the efficiency of the holding tube.

Of the chemical tests used, the sodium nitrite test was preferred. The red precipitate in the nickel chloride–dimethylglyoxime reaction stained the sampling tubes and rack which, in subsequent tests, might give a false impression of colour. This was not so for the sodium nitrite–Griess-Ilosvay test.

The salt conductivity test was more easily adapted to varying conditions than the chemical test and it was not more difficult to operate. The reagents for the chemical test required greater care and time in preparation and handling than the salt solution for the salt conductivity test; the salt conductivity test could be done in much less time than the chemical test.

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A TEST FOR THE DETECTION OF LACTIC BACTERIOPHAGE USING 2,3,5-TRIPHENYLTETRAZOLIUM CHLORIDE [1]

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SUMMARY

In this study, 2,3,5-triphenyltetrazolium chloride (TTC) was used to measure inhibition of lactic cultures by bacteriophage. The procedure follows: Place 8.7 ml. of 10% sterile reconstituted nonfat dry milk (NDM) in each of two sterile test tubes. To each tube add 0.2 ml. of a lactic culture diluted 1:1 with sterile 10% NDM. Add 1 ml. of serially diluted phage filtrate to one tube. To the other tube, remaining as the control, add 1 ml. of sterile distilled water. Place sterile rubber stoppers in the tubes and invert twice to mix the contents. Incubate the tests for 4 hr. at 32° C. in a water bath. Add 0.1 ml. of 1:25 aqueous solution of TTC per tube. Invert the tubes twice and incubate again for 30 min. at 32° C. Compare the color in the tube with added filtrate to the control tube. If less color is developed than in the control, inhibition has occurred. Results of this test are available in 4.5 hr. Its sensitivity for bacteriophage is comparable, and in some cases better, than a litmus milk test *(8)* requiring from 12 to 16 hr. With mixed-strain lactic starter cultures, the TTC test indicates some cases of phage inhibition in 4.5 hr. that are not evident after extended incubation in the litmus milk test. By using unheated filtrate and filtrate heated to 80° C. for 10 min., this test can be used to distinguish between inhibition of lactic cultures caused by heat-stable inhibitors and bacteriophage.

The economic losses caused by bacteriophage in the production of cultured dairy products are well-known. Reviews by Babel *(2)*, Elliker *(3)*, and Whitehead *(9)* cover the subject extensively.

Two techniques have been used to isolate or detect bacteriophage: counting plaques on solid or semisolid media inoculated with serially diluted phage filtrates and susceptible cells; and changes in acidity or oxidation-reduction potential in broths or milk media containing the test organisms and phage filtrates.

The TTC test, an effective indicator of bacterial growth *(7)*, was used in this study to detect bacteriophage action on lactic organisms. It was believed that the TTC test might be applicable for checking sensitivity of single lactic strains to phage strains, and for testing for phage as a cause of starter failure in making cultured dairy products. A litmus milk method has been reported as a reliable method of estimating phage concentrations *(8)*. This method was used as a standard of comparison for adaptation of the TTC test for phage detection.

METHODS

The cultures used in this study were obtained from two sources: the single strain *Streptococcus lactis* cultures from E. B. Collins, University of California, Davis, and the mixed-strain lactic cultures from Chris Hansen's Laboratory, Milwaukee, Wisconsin.

Received for publication December 13, 1957.

[1] Published with the approval of the Director of the Wisconsin Agricultural Experiment Station.

[2] Present address: University of Florida, Gainesville, Florida.

All cultures were propagated in a 10% reconstituted nonfat dry milk medium sterilized at 121° C. for 15 min. in the autoclave. The cultures were transferred daily to retain optimum activity. *S. lactis* single strains were incubated for from 8 to 10 hr. at 32° C. The mixed lactic cultures were used for inoculum after incubation for from 14 to 16 hr. at 22° C.

Bacteriophage strains specific for each of the *S. lactis* single strains also were obtained from E. B. Collins, University of California. Reserve stocks of phage filtrates were prepared, using #03 Selas candles for filtering, to obtain sterility. The phage filtrates were stored at approximately 4° C. They were checked for sterility by adding 1 ml. of the filtrate to 10 ml. of litmus milk and then incubating these samples at 32° C. for 48 hr. After the 48 hr., the litmus milk with added phage filtrate was compared to the control with no phage filtrate added, to measure sterility.

Titers of the phage filtrates were determined by using the litmus milk method proposed by Nelson *(8)*. The determinations were carried out in triplicate at each dilution. The greatest dilution of the phage filtrate with sterile distilled water causing inhibition of growth in at least two of the triplicate determinations was designated the titer. Dilutions of the original phage filtrates up to 10^{-11} were tested.

A TTC solution containing 4 g. of TTC in 96 ml. of sterile distilled water was made; it was stored at 4° C. when not in use. When 0.1 ml. of this solution was added to 10 ml. of medium, a final concentration of approximately 0.4 mg/ml of TTC was established.

A permanent color standard was made by mixing equal parts of aqueous solutions of 0.1% methyl orange and 0.25% safranin and adding the mixture at three levels to 10 ml. of 10% reconstituted sterilized NDM. The three levels of color standard used are shown (Table 1) with the color ratings associated

TABLE 1

Arbitrary color standard for TTC test using 0.1% methyl orange and 0.25% safranin

Milk	Dye mixture	Color rating	Corresponding chip in Munsell Book
(ml.)			
10.0	0.015	(+)	(10.R) 8/4
10.0	0.060	(++)	(5.OR) 7/6
10.0	0.180	(+++)	(5.OR) 6/10

with almost complete inhibition (+), slight inhibition (++), and no inhibition of bacterial growth (+++) during the TTC test. Proper color chips from the Munsell Book of Colors *(5)* were selected and mounted on a card for rapid reference.

The following factors were varied in the TTC test, to find conditions associated with minimum time for test performance, and with sensitivity comparable to the litmus milk method: *(1)* Amount of inoculum; *(2)* concentration of TTC; *(3)* time of adding TTC, and *(4)* total length of time for the test.

After completing preliminary experiments to establish test conditions, the following procedure for the TTC test was used to compare it further with the litmus milk method.

Recommended TTC Method

1. Place 8.7 ml. of 10% sterile reconstituted NDM in each test tube.
2. Bring the tubes' contents up to 32° C. in the water bath.
3. To each tube add 0.2 ml. of a culture diluted 1:1 with sterile 10% NDM.
4. Add 1 ml. of diluted phage filtrate to one tube and 1 ml. of sterile distilled water to the control tube.
5. Place sterile rubber stoppers in tubes and invert twice to mix the contents.
6. Incubate the tubes for 4 hr. at 32° C.
7. Add 0.1 ml. of a 1:25 solution of TTC.
8. Invert twice and incubate for an additional 30 min.
9. Compare the tubes to a control tube with no phage filtrate and rate them according to the color standard.
10. Any tube showing less color development than the control indicates inhibition of bacterial growth.

RESULTS AND DISCUSSION

Factors investigated in determining optimum conditions for the test:

1. *Per cent inoculum.* Since sufficient inoculum is needed for visible color development in a reasonable time, levels of 1, 2, and 4% were used in developing the TTC test. Using a 1% level of inoculum, phage could be detected in higher dilutions of phage filtrates than with 2 or 4% inoculum. With 1% inoculum, adequate growth took place after 4 hr. of incubation at 32° C., to give sufficient color development when TTC was added. Higher levels of inoculum usually coagulated the milk in the control tubes in less than 4 hr. This makes the test ineffective, because added TTC is not reduced; therefore, color does not develop when coagulation has occurred.

2. *Concentration of TTC.* The intensity of color development is in proportion to the numbers of actively metabolizing bacterial cells and the amount of TTC available for reduction. Laxminarayana *(4)* has shown this with several lactic cultures. Neal *(7)* reported that levels of TTC above 1.2 mg/ml caused reduction in color development by an *S. lactis* culture. Mustakallio *(6)* also stated that levels of TTC above 1.0 mg/ml caused inhibition of lactic cultures.

Since the TTC solution is added after a 4-hr. delay and a control is used and the test is read in 30 min., the problem of inhibition is not important. Neal *(7)* used a *Streptococcus thermophilus* culture which is inhibited by levels of TTC lower than those which inhibit a *S. lactis* culture.

The higher the concentration of TTC present, the more the reduction is affected by light *(4)*. Therefore, the lowest amount of TTC giving a good in-

tensity of color should be the most desirable. In this study, levels of TTC of 0.4, 0.8, and 1.2 mg/ml final concentration in the medium were tried.

The color intensity developed was greater using the higher levels of TTC than using 0.4 mg/ml. A control for each level of TTC was always used as a comparison to cancel this error. In some instances, phage was detected at higher filtrate dilutions when using 0.4 mg/ml of TTC than when using 0.8 mg/ml. It appeared that 0.4 mg/ml was a sufficiently high level of TTC to get excellent test results. By keeping TTC concentrations low, light sensitivity was reduced and problems of inhibitions of bacterial growth by the added TTC were lessened.

3. *Time of TTC addition.* Neal *(7)*, in his use of the TTC test for antibiotics with *S. thermophilus* as test culture, found that a delay of 2 hr. before TTC was added to the growing culture gave greatest sensitivity. This same principle was true in the use of the test for bacteriophage detection.

In these experiments, TTC was added at three intervals—at the beginning of incubation, after 2 hr., and after 4 hr. All tests were read at the end of 4.5 hr. incubation. When TTC is added at the beginning of incubation, apparently the organisms cause reduction of the TTC and color development before being attached and lysed by the phage. As a result of this, phage was detected only at dilutions of filtrate up to 10^{-5} when the TTC was added at the start of the incubation period. If a delay of 2 hr. after the initial incubation was allowed before the TTC was added, phage was detected in filtrate dilutions up to 10^{-8}. If a delay of 4 hr. was allowed, phage was detected in dilutions up to 10^{-10}.

This trend was even more evident in the case of one *S. lactis* culture. This culture had a high activity and yielded a phage filtrate with a titer of 8. It had a tendency to overgrow the phage and, when the TTC was added at 0 and 2 hr., phage could be detected only in filtrate dilutions up to 10^{-3} and 10^{-4}. When the TTC was added after 4 hr., the phage had developed and was detected up to dilutions in filtrate of 10^{-7}.

4. *Length of time for the test.* Bacteriophage undergoes a latent period, or lag phase, in its development similar to the lag phase in the growth of bacterial cells. This period of adsorption before development and lysis is an important factor to be considered in tests for phage action. Zehren and Whitehead *(10)* reported 40- to 60-min. latent periods for the six phage strains they tested. Zehren and Whitehead *(10)* also have shown the importance of the burst size, or the number of phage particles, released when a bacterial cell undergoes lysis, on the increase in phage concentrations. Sufficient time must be allowed for the phage to increase in numbers, especially when phage filtrates are diluted out and the concentration of phage particles is low.

From results obtained in this study, it appears that an interval of about 4 hr. of incubation at 32° C. was necessary for the phage development period. For example, at 2.5 hr. time, phage was usually detected only in dilutions of phage filtrate up to 10^{-7}, but at 4.5 hr. the phage in the higher dilutions had increased in numbers and was detected at dilutions up to 10^{-10}, using a 1% inoculum. Increasing the length of time to 6.5 hr. for the test did not increase the sensi-

tivity to any extent. This same trend was true with all cultures used, both single-strain *S. lactis* and mixed-strain lactic cultures.

COMPARISON OF THE TTC AND LITMUS MILK METHODS FOR DETERMINING TITERS OF PHAGE FILTRATES

The litmus milk method has been used by several investigators *(1, 8, 9)* to estimate titers and numbers of particles in serially diluted phage filtrates. An extremely small inoculum, one drop of culture diluted 1:8 in sterile milk medium, was used per 10 ml. of litmus milk and an incubation period of from 12 to 16 hr. was used.

In this study, an attempt was made to obtain comparable results with the TTC method, using a total time of 4.5 hr. for the test. The titers obtained, using four single *S. lactis* strains and two mixed lactic cultures and their respective phages, are shown (Table 2). Results with three of the four *S. lactis* single Strains 4, 5, 7 were identical for both methods. Phage was detectable in dilutions of the various filtrates to 10^{-10}, or a titer of 10 was found. For Strain No. 6, the titer by the TTC method was one dilution less than that by the litmus milk method. This may have been a result of the phage having a longer latent period and the culture having a high activity. As a result of this, it required more time for the phage to develop, cause lysis, and show dominance over the culture.

TABLE 2

Comparison of the sensitivity of the TTC method and the litmus milk method for determining titers of phage filtrates

Culture	Phage filtrate	Highest phage dilution causing inhibition	
		TTC method	Litmus milk method
4	F4	10^{-10}	10^{-10}
5	F5	10^{-10}	10^{-10}
6	F6	10^{-7}	10^{-8}
7	F7	10^{-10}	10^{-10}
3	F4	10^{-3}	No inhibition at 10^{-1}
28	F6	10^{-5}	No inhibition at 10^{-1}

Cultures 3 and 28 were mixed-strain starter cultures, which proved to be sensitive to some degree to the two phages used. Probably only one or, at the most, two of the single strains in each of the mixed strain cultures were sensitive to a phage used. This may be the primary reason why both Cultures 3 and 28 grew over the phage, even at filtrate dilution of 10^{-1}, when the litmus milk method was used. Inhibition by phage filtrate dilutions of 10^{-3} for No. 3 and 10^{-5} for No. 28 was shown when the TTC method of 4.5 hr. of incubation was used. These results are averages of five trials and are presented (Table 2).

This may be a significant point, in that at the present time a recommended procedure to detect phage in cheese plants is to place some susceptible culture and whey from suspected slow vats in sterile milk medium and incubate this at 22° C. for 24 hr. If coagulation occurs, phage is not suspected *(3)*. This may give the cheesemaker a false sense of security, because an active phage may be present which is able to lyse one or two strains in the mixed lactic starter, yet

the remaining unsusceptible strains will produce acid and coagulate the milk in 24 hr. Results (Table 2) show that a susceptible phage with titer of 10^{-5} was not detected after a 12- to 16-hr. incubation period in litmus milk. It was evident after 4.5 hr., using the TTC method, before the unsusceptible *S. lactis* strains developed and caused coagulation.

Results of testing the sensitivity, by both the TTC and litmus milk methods, of 15 single strains of *S. lactis* to two unknown phage filtrates obtained from Wisconsin cheese factories, are shown (Table 3). Usually, the results of the TTC test compared favorably with the results by the litmus milk method. Some strains did show sensitivity to a phage filtrate in the TTC test, but not in the longer litmus milk method. This may be a result either of the single strain being contaminated, or of the organism being only moderately susceptible to the phage and possessing the ability to outgrow it. The ratio between the amount of inoculum and the concentration of phage could be another factor causing some differences between the two tests.

TABLE 3

Results of TTC and litmus milk tests for the sensitivity of 15 single strains of S. lactis to two unknown phage strains obtained from whey

S. lactis strain	TTC method[a]			Litmus milk method[b]		
	Control	Phage XF added	Phage AF added	Control	Phage XF added	Phage AF added
1	3+	−	−	+	−	−
2	3+	2+	−	+	−	−
3	3+	−	1+	+	−	−
4	3+	3+	3+	+	+	+
5	3+	−	1+	+	−	−
6	3+	−	3+	+	−	+
7	3+	1+	3+	+	−	+
8	3+	−	3+	+	−	+
9	3+	−	−	+	−	−
10	3+	−	3+	+	−	+
11	3+	3+	−	+	+	−
12	3+	2+	1+	+	−	−
13	3+	2+	3+	+	+	+
14	3+	2+	1+	+	+	+
15	3+	3+	3+	+	+	+

[a] −, very strong inhibition; 1+, strong inhibition; 2+, mild inhibition, 3+, no inhibition.
[b] +, no inhibition; −, inhibition.

USE OF THE TTC TEST TO DIFFERENTIATE BETWEEN INHIBITION CAUSED BY HEAT-STABLE INHIBITORS AND BACTERIOPAHGE

The test can be a means of determining which of the two inhibitory agents are responsible for a starter failure, by making duplicate determinations. In one determination, use the suspected whey filtrate or milk as obtained; in the other, add the suspected filtrate or milk after heating it to 80° C. for 10 min. When the heated filtrate causes inhibition of the particular starter culture from above, heat-stable inhibitors which are not destroyed at 80° C. for 10 min. are probably responsible for the starter failure. The problem is almost surely bacteriophage when inhibition occurs when the unheated filtrate is used, but not when the heated filtrate or milk is added. By using this test, the cause of starter

failure could be determined and corrective measures started before milk was received on the following day.

ACKNOWLEDGMENTS

This study was supported by the Research Committee of the Graduate School from funds supplied by the Wisconsin Alumni Research Foundation.

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COMPARATIVE ACCURACY OF SMALL LACTOMETERS FOR DETERMINING THE TOTAL SOLIDS OF INDIVIDUAL COWS' MILK [1,2]

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SUMMARY

Milks were obtained monthly from approximately 350 cows of five dairy breeds. Averaged total solids measured with the small Watson lactometer did not differ significantly (except three Brown Swiss milks, 6% fat) from those measured gravimetrically. Correlations between the small Watson lactometer and gravimetric measurements exceeded 0.9 when fat levels were ignored, were lower when the data were grouped by fat percentages, and mostly were statistically significant. Standard errors of estimate ranged from 0.14 to 0.39, indicating errors in total solids equivalent to 0.12% fat by the Babcock method. Field use of the small Watson lactometer can be recommended, provided temperature and reading techniques are controlled.

Average total solids measured by the small Quevenne lactometer differed significantly from the gravimetric measurements in milk from several breed and fat level groups. Correlations and standard errors were of the same magnitude as those obtained with the small Watson lactometer. A significant interaction between methods (gravimetric vs. Quevenne lactometer) and fat levels was obtained when measurements for all breeds were combined, indicating an incapacity of the small Quevenne lactometer and/or the estimation equation to measure accurately total solids at various fat levels. This bias was positive at low fat and negative at high fat levels, indicating that the small Quevenne lactometer is not as accurate as the Watson lactometer.

Lactometric procedures for measuring total solids have been criticized because of inaccuracies and the relatively large quantities of milk required for their use. The recent availability of two lactometers requiring small quantities of milk suggested a study of the measurement of total solids with these lactometers compared with a standard gravimetric method. Such lactometers, if sufficiently accurate, would provide a rapid test for the total solids of individual cows' milk.

This study evaluated two small lactometers as a method for measuring the total solids of milks from individual cows of five dairy breeds.

EXPERIMENTAL METHODS

Samples. Four-ounce milk samples were collected monthly by the DHIA supervisor, from the Michigan State University dairy herds (five breeds) and seven cooperator herds (one Guernsey, five Holstein, and one Jersey herd). The distribution of cows by breed was eight Ayrshire, 30 Brown Swiss, 68 Guernsey, 180 Holstein, and 62 Jersey.

Measurements. Fat values for each sample were obtained from official DHIA records. Total solids measured by a gravimetric method on the same milk sample

Received for publication January 30, 1958.

[1] Journal Article No. 2196 from the Michigan Agricultural Experiment Station.

[2] These studies were supported in part by the Dairy Cattle Research Branch of the Agricultural Research Service, USDA.

were considered as reference values. The two lactometers used in this study to estimate total solids were: (a) the small Watson-pattern lactometer, read at 102° F., approximately 6 in. long, and requiring about 4 oz. of sample *(3)*; and *(b)* a small Quevenne lactometer of the type in official use in the Netherlands, to be read at 68° F. after previous warming to 102° F., approximately 8 in. long, and requiring about 4 oz. of sample *(2)*. Estimation of total solids from the lactometers noted above was computed as follows:

$$\text{Lactometer } (a) \quad \text{T.S.} = 1.33\,\text{F} + \frac{273\,\text{L}}{\text{L} + 1000} - 0.40 \qquad (3)$$

$$\text{Lactometer } (b) \quad \text{T.S.} = 1.23\,\text{F} + \frac{260\,(\text{L} + 1.8)}{\text{L} + 1000} \qquad (2)^{3}$$

F = Babcock fat %. L = Lactometer reading in degrees.

TABLE 1

Total solids measured by the small Watson lactometer and gravimetric methods, by breeds and fat levels (February through June, 1957)

Breed	Fat levels (%)							
	<3	3	4	5	6	7	8	All
Holstein								
No. samples	11	469	191	12	1	.....	.....	684
Av. fat	2.7	3.5	4.3	5.2	6.0	.....	.....	3.8
Av. T.S. (G)[a]	11.06	12.13	13.15	14.21	15.92	.....	.....	12.44
Av. T.S. (L)[a]	11.17	12.14	13.15	14.33	15.61	.....	.....	12.45
Guernsey								
No. samples	1	16	188	94	24	2	.....	325
Av. fat	2.4	3.7	4.4	5.3	6.2	7.4	.....	4.8
Av. T.S. (G)	10.43	12.48	13.56	14.63	15.78	16.95	.....	13.99
Av. T.S. (L)	10.36	12.52	13.48	14.61	15.86	16.66	.....	13.94
Jersey								
No. samples	.....	.....	46	123	54	10	.....	233
Av. fat	.....	.....	4.6	5.5	6.3	7.2	.....	5.6
Av. T.S. (G)	.....	.....	14.01	15.05	16.08	17.01	.....	15.17
Av. T.S. (L)	.....	.....	13.97	15.07	16.16	17.21	.....	15.20
Brown Swiss								
No. samples	1	44	51	7	3	.....	.....	106
Av. fat	2.7	3.6	4.4	5.3	6.2	.....	.....	4.2
Av. T.S. (G)	11.09	12.60	13.59	15.04	16.35	.....	.....	13.33
Av. T.S. (L)	10.88	12.52	13.72	15.05	15.60[b]	.....	.....	13.33
Ayrshire								
No. samples	.....	17	17	2	.....	.....	1	37
Av. fat	.....	3.6	4.3	5.2	.....	.....	8.5	4.2
Av. T.S. (G)	.....	12.51	13.42	14.13	.....	.....	18.65	13.18
Av. T.S. (L)	.....	12.44	13.18	13.86	.....	.....	17.69	13.00
All Breeds								
No. samples	13	546	493	238	82	12	1	1,385
Av. fat	2.7	3.6	4.4	5.4	6.3	7.3	8.5	4.4
Av. T.S. (G)	11.02	12.19	13.44	14.83	16.00	17.00	18.65	13.35
Av. T.S. (L)	11.08	12.19	13.41	14.84	16.05	17.12	17.69	13.34

[a] Average T.S. (G) and (L) refer to average total solids obtained by the gravimetric and lactometric methods, respectively.

[b] Significantly different from average total solids measured gravimetrically at P = 0.05.

[3] Equivalent to the reported estimation equations:

$$D = 1.23\,V + 2.6 \times 100\,(d^{20} - 0.9982)/d^{20}$$

where V and d^{20} refer to fat per cent and density of the milk at 20° C., respectively.

RESULTS AND DISCUSSION

Since fat levels and breed characteristics are traditionally considered as the major causes of variations in total solids, these two factors were considered in comparing the lactometric procedures with the gravimetric measurements.

Small Watson lactometer. The results of the total solids measurements using the small Watson lactometer and the gravimetric method are presented (Tables 1 and 2). The average lactometer measurements were not significantly different from the gravimetric measurements, except in three Brown Swiss samples at the 6% fat level, when fat levels and/or breeds were considered. The correlations between total solids measured lactometrically and gravimetrically were statistically significant, except for Guernsey samples at 6% and Brown Swiss samples at 5 and 6% fat levels. These correlations ranged from −1.0 to 1.0 for fat levels within breeds, from 0.90 to 0.94 for breeds when fat levels were ignored, and from 0.54 to 0.85 for fat levels when breeds were ignored. A correlation of 0.96 was obtained when breeds and fat levels were ignored. Standard errors of estimate ranged from 0.14 to 0.39 for fat levels within breeds, from 0.28 to 0.40 for breeds when fat levels were ignored, and from 0.23 to 0.39 for fat levels when breeds were ignored. A standard error of estimate of 0.36 was obtained when breeds and fat levels were ignored.

Watson *(3)*, using his large lactometer, reported 59 tests on individual cows of four dairy breeds with an average deviation of +0.05% and a standard error of estimate of 0.13. In a previous study, the authors *(1)* reported an average deviation of +0.20% and a standard error of estimate of 0.40 when a lactometer of the type employed by Watson was used on 1,454 Jersey samples. Correspond-

TABLE 2

Correlations and standard errors between total solids measured gravimetrically and with the small Watson lactometer, by breeds and fat levels (February through June, 1957)

Breed	Fat levels (%)						
	<3	3	4	5	6	7	All
Holstein							
r[a]	0.73	0.84	0.82	0.83	.....	.....	0.92
SE[b]	0.22	0.23	0.27	0.20	.....	.....	0.28
Guernsey							
r	.....	0.63	0.71	0.83	0.32[c]	1.00	0.90
SE	.....	0.25	0.31	0.24	0.26	.....	0.40
Jersey							
r	.....	.....	0.67	0.74	0.65	0.87	0.91
SE	.....	.....	0.27	0.30	0.39	0.28	0.39
Brown Swiss							
r	.....	0.84	0.71	0.41[c]	−0.96[c]	.....	0.92
SE	.....	0.26	0.33	0.30	0.14	.....	0.39
Ayrshire							
r	.....	0.92	0.66	−1.00	.....	.....	0.94
SE	.....	0.18	0.39	.....	.....	.....	0.36
All Breeds							
r	0.79	0.85	0.78	0.81	0.54	0.82	0.96
SE	0.23	0.23	0.32	0.29	0.39	0.32	0.36

[a] r refers to the correlation between total solids measured gravimetrically and lactometrically.

[b] SE refers to the standard error of estimate.

[c] Not significantly different from zero at P = 0.05.

ing values of +0.01% and 0.36 are indicated for the small Watson lactometer (Tables 1 and 2). Under circumstances similar to those in the study reported here, 95% of the calculated total solids would be expected to fall within −0.01 ±0.72%, or from −0.73 to +0.71% total solids from the gravimetric values. At the average fat levels noted, an error of similar magnitude in fat test would be 0.24%.

Analyses of variance by breeds and for all breeds combined showed no significant differences between the lactometric and gravimetric methods for measuring total solids. Highly significant differences in total solids for fat levels were as expected. The absence of significant interactions between methods and fat levels indicated that different fat levels did not affect the relations between the two methods of measurement. The error mean squares ranged from 0.25 to 0.33.

These data indicate that the small Watson lactometer is useful for measuring the total solids of individual cows' milk. The value of the technique for field use is dependent on standard procedures, such as temperature control and reading techniques.

Small Quevenne lactometer. The results of the total solids measurements,

TABLE 3

Total solids measured by the small Quevenne lactometer and gravimetric methods, by breeds and fat levels (October, 1956, through June, 1957)

Breed	Fat levels (%)						
	<3	3	4	5	6	7	All
Holstein							
No. samples	13	508	226	12	.....	.....	759
Av. fat	2.7	3.5	4.3	5.2	.....	.....	3.8
Av. T.S. (G)[a]	11.22	12.14	13.17	14.21	.....	.....	12.47
Av. T.S. (L)[a]	11.40	12.21[b]	13.19	14.29	.....	.....	12.52
Guernsey							
No. samples	1	18	198	107	32	2	358
Av. fat	2.4	3.7	4.4	5.3	6.3	7.4	4.8
Av. T.S. (G)	10.43	12.40	13.56	14.68	15.86	16.95	14.05
Av. T.S. (L)	10.49	12.51	13.51	14.62	15.78	16.42[b]	14.00
Jersey							
No samples	.....	1	49	153	95	14	312
Av. fat	.....	3.9	4.6	5.5	6.3	7.2	5.7
Av. T.S. (G)	.....	11.22	13.96	15.09	16.13	17.11	15.30
Av. T.S. (L)	.....	11.72	13.94	15.00	15.96[b]	16.85	15.20
Brown Swiss							
No. samples	1	60	94	21	3	.....	179
Av. fat	2.8	3.7	4.4	5.2	6.3	.....	4.3
Av. T.S. (G)	11.26	12.49	13.72	14.96	16.07	.....	13.48
Av. T.S. (L)	11.35	12.50	13.67	14.81	15.60	.....	13.43
Ayrshire							
No.samples	.....	22	25	2	.....	.....	49
Av. fat	.....	3.6	4.4	5.0	.....	.....	4.1
Av. T.S. (G)	.....	12.56	13.38	14.13	.....	.....	13.04
Av. T.S. (L)	.....	12.56	13.35	14.09	.....	.....	13.03
All Breeds							
No. samples	15	609	592	295	130	16	1,657
Av. fat	2.7	3.6	4.4	5.4	6.3	7.2	4.4
Av. T.S. (G)	11.17	12.20	13.47	14.89	16.06	17.08	13.47
Av. T.S. (L)	11.34	12.26[b]	13.44	14.82	15.91[b]	16.79	13.46

[a] Average T.S. (G) and (L) refer to average total solids obtained by the gravimetric and lactometric methods, respectively.

[b] Significantly different from average total solids measured gravimetrically at P = 0.05.

using this lactometer and the gravimetric method, are presented (Tables 3 and 4). In several breed and fat groups, the average total solids measured by the lactometer were significantly different from those measured gravimetrically. The direction of the differences indicated that total solids measured lactometrically were higher than the gravimetric measurements at the low fat level (Holsteins and "All Breeds" at 3% fat) and lower at the high fat levels (Jerseys, Guernseys, and "All Breeds" at the 6 and 7% fat levels). Several of the significant differences were in groups with numerous observations, which indicated a real defect in the ability of this lactometer and/or the estimation equation to measure total solids.

All correlations presented (Table 4) were significant statistically, except for Brown Swiss samples at the 6% fat level. These correlations ranged from −0.13 to 1.00 for fat levels within breeds, from 0.91 to 0.96 for breeds when fat levels were ignored, and from 0.58 to 0.88 for fat levels when breeds were ignored. A correlation of 0.97 was obtained when fat levels and breeds were ignored. The corresponding standard errors of estimate ranged from 0.18 to 0.45, from 0.24 to 0.38, and from 0.20 to 0.35, respectively. A standard error of estimate of 0.32 was obtained when breeds and fat levels were ignored.

Analyses of variance by breeds and for all breeds combined showed significant differences between the lactometric and gravimetric methods for measuring total solids in the Holstein and Jersey groups. Again, highly significant differences between fat levels indicated that total solids of milk differed with fat levels. A significant interaction obtained between methods and fat levels for the data for all breeds combined indicated that the relations between the two methods differed

TABLE 4

Correlations and standard errors between total solids measured gravimetrically and with the small Quevenne lactometer, by breeds and fat levels (October, 1956, through June, 1957)

Breed	Fat levels (%)						
	<3	3	4	5	6	7	All
Holstein							
r[a]	0.83	0.88	0.85	0.80	.....	.....	0.93
SE[b]	0.22	0.19	0.24	0.20	.....	.....	0.25
Guernsey							
r	.....	0.80	0.73	0.86	0.40	1.00	0.92
SE	.....	0.32	0.30	0.22	0.24	.....	0.37
Jersey							
r	.....	.....	0.79	0.69	0.61	0.81	0.91
SE	.....	.....	0.29	0.32	0.37	0.26	0.38
Brown Swiss							
r	.....	0.91	0.84	0.80	−0.13[c]	.....	0.96
SE	.....	0.18	0.25	0.22	0.45	.....	0.27
Ayrshire							
r	.....	0.86	0.85	1.00	.....	.....	0.93
SE	.....	0.23	0.22	.....	.....	.....	0.24
All Breeds							
r	0.87	0.88	0.83	0.80	0.58	0.80	0.97
SE	0.22	0.20	0.28	0.28	0.35	0.28	0.32

[a] r refers to the correlation between total solids measured gravimetrically and lactometrically.

[b] SE refers to the standard error of estimate.

[c] Not significantly different from zero at P = 0.05.

In view of the rather wide diversity, both in methods of treatment and in the results reported, a study was undertaken in goats to compare the effectiveness for inducing lactation of stilbestrol and progesterone-stilbestrol combinations in several different ratios that were varied over a wide range. To circumvent the necessity of pellet implantation, or daily injection of an oil solution, the hormones were prepared as a macrocrystalline aqueous suspension that has a sustained effect and yet is well absorbed.

METHODS AND PROCEDURES

Eighteen dairy goats including Rock Alpines, Saanens, Toggenburgs, and Crossbreds were divided into four treatment groups that were matched as nearly as possible for age and breed (Table 1).

TABLE 1
Experimental design

Group	Goat No.	Breed	Age when experiment started	Stilbestrol	Progesterone
				(weekly)	
I	4	Saanen	2 yr.	1.75 mg.(11 wk.)	None
	19	Toggenburg-Nubian	8 mo.	1.75 mg.(11 wk.)	None
	7[a]	Nubian	4 yr.	1.75 mg.(11 wk.)	None
	16	Rock Alpine	8 mo.	1.75 mg.(11 wk.)	None
II	6	Toggenburg-Nubian	5 yr.	1.75 mg.(11 wk.)	70 mg.(9 wk.)
	18	Saanen	8 mo.	1.75 mg.(11 wk.)	70 mg.(9 wk.)
	8	Nubian	4 yr.	1.75 mg.(11 wk.)	70 mg.(9 wk.)
	21	Saanen	8 mo.	1.75 mg.(11 wk.)	70 mg.(9 wk.)
III	14	Rock Alpine	1 yr. 8 mo.	0.875 mg.(11 wk.)	70 mg.(9 wk.)
	24	Saanen	8 mo.	0.875 mg.(11 wk.)	70 mg.(9 wk.)
	15	Rock Alpine	1 yr. 8 mo.	0.875 mg.(11 wk.)	70 mg.(9 wk.)
	5	Saanen	8 mo.	0.875 mg.(11 wk.)	70 mg.(9 wk.)
IV	9	Toggenburg-R. Alp.	5 yr.	0.07 mg.(11 wk.)	70 mg.(9 wk.)
	22[a]	Saanen	8 mo.	0.07 mg.(11 wk.)	70 mg.(9 wk.)
	12	Nubian	2 yr.	0.07 mg.(11 wk.)	70 mg.(9 wk.)
	23[a]	Saanen	8 mo.	0.07 mg.(11 wk.)	70 mg.(9 wk.)
	10	Saanen-R. Alp.	1 yr. 8 mo.	0.07 mg.(11 wk.)	70 mg.(9 wk.)
	11	Toggenburg	1 yr. 8 mo.	0.07 mg.(11 wk.)	70 mg.(9 wk.)

[a] Animal not responding to initial treatment.

The macrocrystalline steroids to be administered were ground in a Potter-Elvehjem homogenizer with a 0.3% starch emulsion, a preparation which supplies a prolonged exposure of approximately 2 wk. [Nellor and Cole *(12)*]. The amount of progesterone administered (70 mg. per week) was the lower limit of the amount necessary *(10)* to maintain pregnancy in ovariectomized goats. The duration of treatment (9–11 wk.) corresponds to approximately one-half of the normal gestation period in the goat.

Two weekly injections of stilbestrol terminated each series of weekly injections on the supposition that this would facilitate the release of lactogenic hormone from the pituitary, as suggested by earlier work of Meites *et al. (9)*. Benson *et al. (1)* reported that estrogen priming, subsequent to progesterone-

estrogen treatment, had no beneficial effects on induced lactations in dairy goats. Although the progesterone-estrogen ratios in their study were wide, the amount of estrogen used was high. This might preclude the use of an estrogen primer, because sufficient estrogen may already have been present in the circulation.

Group I (Table 1) received 1.75 mg. of stilbestrol weekly for 11 wk. Group II received 1.75 mg. of stilbestrol and 70 mg. of progesterone; Group III received 0.875 mg. stilbestrol and 70 mg. progesterone, and Group IV received 0.07 mg. of stilbestrol and 70 mg. of progesterone, in a single subcutaneous injection once weekly for 9 wk. This represents a progesterone-estrogen empirical ratio of 40:1, 80:1, and 1,000:1, respectively. Groups II, III, and IV received two additional injections of 1.75 mg. of stilbestrol during the tenth and 11th wk. of the trial. The initial injection was given November 18, 1955.

Eight does were retreated 3 mo. from the start of lactation, to determine the effects of additional hormone therapy on animals failing to respond to the initial treatment and also on the existing lactation of does that had responded to the treatment.

Twice-a-day milking was initiated in Groups I and II when udder distention was noted during the tenth wk. of treatment, and in Groups III and IV one wk. later, one day before the last stilbestrol injection.

The goats were fed second-cutting alfalfa hay ad libitum, received 1.5 lb. of concentrate (16.0% protein) daily during the milking period, and were placed on pasture 10 wk. from the start of milking. Twice-a-day milking was continued for 33 wk. unless the doe did not respond to the treatment, in which case milking was discontinued. Milking was terminated on October 15, 1956, in all does still lactating at that time.

The level of stilbestrol injected did not appear excessive, since only one of the stilbestrol-treated does (No. 16) showed estrus during the treatment. Slight vulvar stimulation was detected in all treated groups by the end of the 3rd wk. of treatment, a phenomenon more evident in the kids than in the aged animals. Weekly palpation of the udder region revealed some development of the udder pad of all treated groups as early as the end of the 4th wk. of treatment, although comparison of the degree of development was not feasible by this method.

The presence of a serous fluid was noted in the teats of a few goats from each group by the 5th wk. of treatment, but obvious distention of the gland with fluids was not apparent in the 80:1 or 1,000:1 groups during the combined progesterone-estrogen treatment.

The most distinct difference in lactational response is found in comparing Groups I (stilbestrol only) with the three groups receiving progesterone-stilbestrol combinations (Figure 1). Only three of the four animals in Group I came into lactation. Peak production was reached later and was generally at a lower level than in the other groups. This is in accord with earlier findings *(11)*, that both progesterone and estrogen are needed for normal mammary growth. However, in comparing the results for Groups II, III, and IV, little evidence is found for an optimal ratio between progesterone and estrogen at the levels and

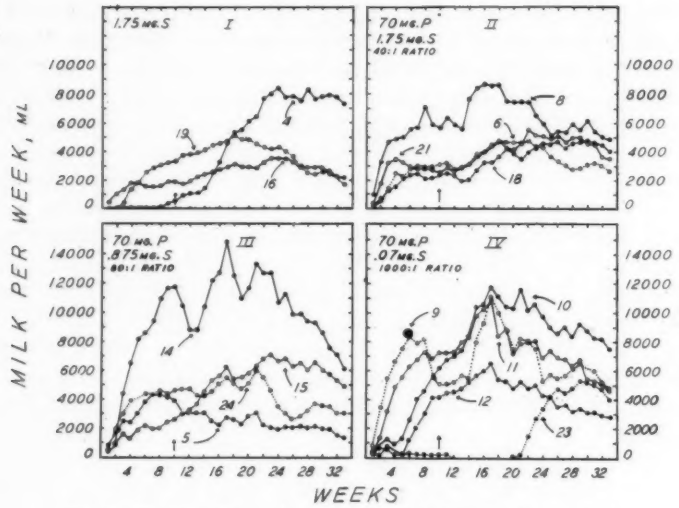

FIG. 1. Weekly milk production induced by hormonal treatment.

in the proportions employed in these trials. All animals in Groups II and III came into lactation. Goat No. 14 (Group III) had the highest production in the entire experiment, yet the remaining animals in this group have records very similar to the three lowest producers in Group II. In the first three groups all of the 8-mo.-old kids responded to the initial treatment with some degree of lactation. In Group IV only the four sexually mature goats responded, no appreciable lactation being obtained in the 8-mo.-old kids. Consequently, the tendency toward higher milk production in Group IV as a whole can probably be accounted for by the disparity in ages.

Retreatment of three of the goats that failed to respond initially (No. 7, Group I, and No. 22 and 23, Group IV) with 0.875 mg. of stilbestrol and 70 mg. of progesterone weekly for 7 wk., resulted in only one doe (No. 23) responding and reaching a production level comparable to that of other responsive animals. Similar retreatment of does already in lactation (No. 16, Group I; No. 6 and 21, Group II, and No. 5 and 15, Group III) caused no apparent inhibition or enhancement of milk production, either during the retreatment or subsequent to it.

In Table 2, the milk yields for the experimental lactations of 12 of the goats that kidded the following year are compared with their normal lactations. Age corrections have not been employed, because factors are not available that would apply to our conditions. From this comparison, it is again apparent that sexually immature animals are distinctly less responsive to the treatments given than are mature goats. Five kids that were approximately 8 mo. of age at the beginning of treatment had 6-mo. experimental milk yields ranging from 20.3 to 54.5% of their normal, and averaged 33%. Seven older animals averaged 62.1% of normal, ranging from 48.8 to 86.3%.

TABLE 2

Induced versus normal[a] milk production

Group	Goat No.	Age		Max. monthly production					6-month production (183 days)[b]		
				Induced		Normal			Induced	Normal	
		Yr.	Mo.	Month	Pounds	Month	Pounds	Normal	Pounds	Pounds	Normal
								(%)			*(%)*
I	4	2		8	77.9	3	130.5	59.7	361.7	616.7	58.7
	19		8	5	47.0	3	215.8	21.8	220.4	889.3	24.8
	16		8	6	33.1	2	144.9	22.8	159.8	612.9	26.1
II	18		8	8	47.0	2	123.0	38.2	217.3	550.7	39.5
	8	4		5	97.2	2	117.9	82.4	408.2	554.8	73.6
III	14	1	8	5	126.4	2	169.3	74.7	640.9	742.7	86.3
	24		8	5	53.0	2	123.3	43.0	259.7	476.5	54.5
	15	1	8	5	65.9	2	136.1	48.4	305.4	626.3	48.8
	5		8	2	41.3	2	160.4	25.7	169.7	834.4	20.3
IV	12	2		4	57.2	2	167.7	34.1	259.0	621.0	41.7
	23		8	5	49.7	2	122.8	40.5	.....	.....	.....
	10	1	8	5	107.6	2	125.3	85.9	517.7	600.2	86.3
	11	1	8	4	92.2	2	149.2	61.8	446.0	692.7	64.4

[a] Obtained from subsequent lactation following pregnancy. Animals not included were not bred following the induced lactation.

[b] Six successive months of highest production.

Two of these animals (No. 14 and No. 10) approached their normal lactations closely, when the difference in age between successive records is considered. However, they were from two different groups, receiving widely different ratios of progesterone to stilbestrol. This raises the question whether a dosage level and combination can be found that will be optimal for all animals, or whether there is too much variability in hormone requirement and responsiveness between animals for this approach to be feasible. It will be necessary to test further hormone combinations and dosage levels before this question can be answered.

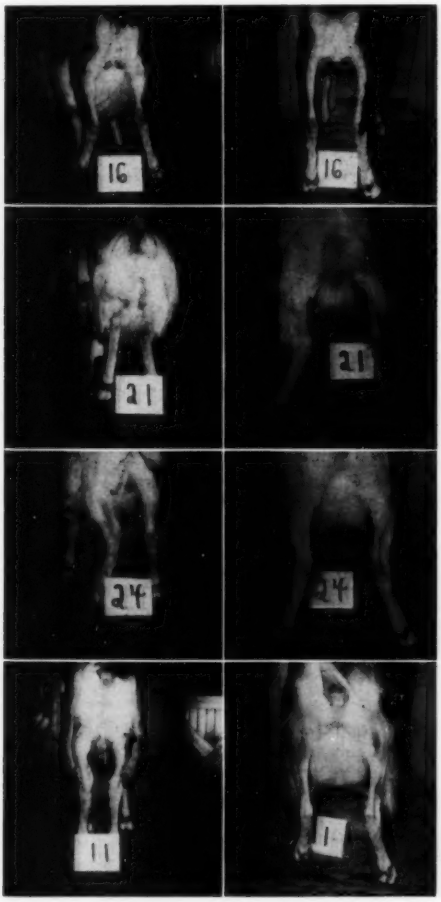

FIG. 2. Mammary development induced by hormonal treatment. Photographs at left at time of initial treatment. Photographs at right 12 wk. later. No. 16, 1.75 mg. stilbestrol per week; No. 21, 24, and 11 each received 70 mg. of progesterone weekly, and 1.75, 0.875, and 0.07 mg. stilbestrol, respectively.

It is of interest to compare visually the udder development in those does not having had a lactation previous to treatment, although it is recognized that this type of comparison has serious limitations. The gross udder development in kids No. 16, 21, and 24 and a yearling doe, No. 11 (Figure 2), is typical of their respective groups.

ACKNOWLEDGMENT

The authors are indebted to Mr. Horst Kaczkofsky for technical assistance with the experiments reported herein.

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INFLUENCE OF STAGE OF MATURITY AND OF GROUND SNAP CORN OR SODIUM METABISULFITE AS PRESERVATIVES ON THE FEEDING VALUE OF OAT SILAGE [1]

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SUMMARY

Fall-sown oats, harvested and stored in 20-ton upright silos, were evaluated with milking cows. In 1956, two silos were filled with oats cut at the prebloom stage, one at milk and one at dough stage. The 1957 silages included two silages cut at the boot stage, two cut at the prebloom stage, and one cut at the milk stage. Ground snap corn and sodium metabisulfite were compared as preservatives. Optimum dry matter preservation was achieved with the ground snap corn, with oats cut at the prebloom stage. In six silos where direct comparison was possible, the ground snap corn preserved 13% more dry matter than the sodium metabisulfite. Dry matter intake was only slightly influenced by the preservative and increased with stage of maturity through the milk stage in 1956 and prebloom stage in 1957. Persistency of milk production over a 28-day period was used as the criteria of feeding value. Only the oats cut at the boot stage were satisfactory for milk production when preserved with sodium metabisulfite, but all silages preserved with ground snap corn resulted in normal milk production.

In recent years, there has been a surge of interest in oat silage as a feed for dairy cows. This interest has resulted from low yields of corn during dry summers and a need for a spring-harvested silage for summer feeding. Few references were found in the literature relative to the influence of stage of maturity, and none on the value of ground snap corn and sodium metabisulfite as preservatives for oat silage.

Meyer *et al. (5)* harvested oat forage from the same field at seven stages of maturity and fed the hay to growing sheep. The highest nutritive value was found in the jointing stage, but the greatest production of nutrients per acre was from the 18% flower stage.

Thurman *et al. (6)* harvested oats at six stages of maturity and determined digestibility coefficients with steers. They concluded that oats should be cut in the boot-to-milk stage for high-quality silage.

Bratzler *et al. (2)* reported marked improvement in nutrient preservation from the use of sodium metabisulfite. Variability between control silages was observed by Gordon *et al. (4)* to be the primary factor influencing the apparent benefit obtained from preservatives. Gordon *et al. (3)* and Bohstedt *et al. (1)* reported improved nutrient preservation with corn meal.

EXPERIMENTAL PROCEDURE

Fall-sown oats were harvested at various stages of maturity and stored in 20-ton upright silos. Each load of silage was weighed before unloading, and

Received for publication December 31, 1957.

[1] Georgia Experiment Station Journal Series, N. S. 327.

the proper amount of preservative was carefully weighed and added. Eight pounds of sodium metabisulfite or 200 pounds of ground snap corn was used per ton of fresh oat forage. The ground snap corn was spread over the silage on the truck and the sodium metabisulfite was added at the blower. Dates of cutting, yield-per-acre, dry matter content, and maximum temperature recorded at the center of each silo are presented (Table 1). The silage cut in 1956 was from an excellent oat crop, whereas the 1957 silage represents a year when crown rust damaged the crop.

In 1956, the three stages at which oat forage was ensiled were the prebloom, milk, and dough; in 1957, the three stages were the boot, prebloom, and milk. For convenience, the silages were numbered consecutively; the last number indicates year of harvest.

Silages 4–6 and 5–6, 6–7 and 7–7, and 9–7 and 10–7 were made by filling silos with alternate loads of forage. All other silages were harvested as single silos. To simulate farm conditions, each silo was filled at the approximate rate of filling expected in making silage in 100-ton silos.

The silos were opened and silages fed as needed. The silage was weighed out of the silo and samples were collected three times each week for dry matter, pH, and other analysis. Samples were frozen, and composited when the silo was empty, for the usual "proximate" analysis. Analytical methods were those recommended at a conference of silage investigators *(6)*.

Feeding value of the silages was evaluated in 28-day feeding trials with four or five milking cows. The cows were fed silage free choice and grain was fed at about 1 : 6. In some instances, it was necessary to feed cottonseed meal because of the low protein content of the silage. Hay was fed with the first two silos, but was discontinued to enable the remaining silos to be fed according to recommendations of the silage conference *(6)* referred to above. Silages 1–6 and 8–7 are included for reference only. Silage 1–6 was fed with temporary winter

TABLE 1

Date of cutting, yield per acre, dry matter content, and maximum silo temperature of the silages

Stage of maturity and silage No.	Date of cutting	Yield per acre	Dry matter of forage	Maximum temperature in silo
		(tons)	*(%)*	*(° F.)*
Dough				
1–6[a]	5–9–56	15.0	32	85
Milk				
2–6	5–1–56	16.0	29	84
3–7	4–29–57	8.0	35	94
Prebloom				
4–6	4–20–56	15.0	25	78
5–6	4–20–56	15.0	25	90
6–7	4–24–57	9.5	28	95
7–7	4–24–57	9.5	28	100
8–7	4–22–57	9.0	24	100
Boot				
9–7	4–10–57	9.7	24	72
10–7	4–10 57	9.7	24	85

[a] Second number indicates year in which silage was made.

pasture, and Silage 8–7 was stored in a trench silo, but represents an untreated silage. Milk-production persistency during the 28-day feeding trial was considered the chief criteria of feeding value.

RESULTS AND DISCUSSION

Preservation of nutrients. The composition of the silages is shown in Table 2, and the preservation of nutrients, in Table 3. On the basis of physical characteristics such as odor, color, fermentation type, and pH, all of the silages would have been scored as good to excellent on the score-cards suggested by the silage conference *(6)*. (Stage of maturity was ignored in scoring, because it was one of the factors under study.) With the exception of Silages 4–6 and 9–7, the temperatures were within the optimum range of 80–100° F. In general, the

TABLE 2

Chemical composition of the silages (dry matter basis)

Stage of maturity and preservative	Silage No.	Crude protein	Ether extract	Crude fiber	N.F.E.	Ash	pH	Dry matter
		(%)						
Dough								
$Na_2S_2O_5$	1–6[a]	7.58	4.22	35.02	47.33	5.85	4.1	27
Milk								
$Na_2S_2O_5$	2–6	9.20	5.66	20.90	46.94	7.30	4.1	27
None	3–7	8.65	2.94	36.99	46.26	5.16	4.0	29
Prebloom								
$Na_2S_2O_5$	4–6	10.71	3.76	29.11	47.27	9.15	4.3	25
Snap Corn	5–6	9.31	8.03	23.59	54.41	4.66	3.7	30
$Na_2S_2O_5$	6–7	10.12	3.24	36.36	44.45	5.83	4.1	25
Snap Corn	7–7	10.30	3.59	29.87	52.06	4.18	3.9	29
None	8–7	8.24	2.92	37.56	45.94	5.34	3.8	24
Boot								
$Na_2S_2O_5$	9–7	10.47	4.38	35.58	43.43	6.14	4.1	19
Snap Corn	10–7	11.93	4.23	27.00	51.55	5.29	3.8	28

[a] Second number indicates year in which silage was made.

chemical composition followed the expected changes with stage of maturity. Crude protein was lower than desired, although all silages were higher in crude protein in the 1957 crop than in the 1956 crop. The lower per-acre yields may have influenced this fraction. The usual changes in chemical composition—increases in crude fiber and ether extract and a decrease in NFE—were observed. Despite the care taken in weighing the forage and silage, there were occasional small losses due to wind and accidents. It was felt, however, that the forage losses were less than 100 lb. per silo.

The data (Table 3) also follow a general pattern relative to preservation of total nutrients. Within a preservative treatment, dry matter preservation increased with increasing stage of maturity until the dough stage. The lower percentage of preservation in the dough stage was due to heavy losses from spoilage resulting from poor compaction. This loss took the form of rotted silage near the walls of the silo and was difficult to weigh accurately, because the exact point of separation between good and spoiled silage was not clear. The

TABLE 3

Per cent of stored nutrients preserved as good silage

Stage of maturity and preservative	Silage No.	Fresh material	Per cent of stored constituents preserved					
			Dry matter	Crude protein	Ether extract	Crude fiber	N.F.E.	Ash
		(%)						
Dough								
$Na_2S_2O_5$	1–6[a]	91.6	76.8	83.0	101.6	86.4	66.9	100.0
Milk								
$Na_2S_2O_5$	2–6	88.8	82.5	93.5	157.2	83.6	71.6	133.5
None	3–7	95.5	79.0	88.7	104.8	90.4	68.8	77.4
Prebloom								
$Na_2S_2O_5$	4–6	85.7	86.4	108.5	116.0	98.2	70.6	150.0
Snap Corn	5–6	87.3	94.0	100.8	263.0	103.5	81.6	99.5
$Na_2S_2O_5$	6–7	84.2	75.9	74.2	109.5	90.9	64.3	97.4
Snap Corn	7–7	89.6	84.9	88.3	130.8	87.7	81.2	80.1
None	8–7	.....	.....	.....	.....	.....	.....	.....
Boot								
$Na_2S_2O_5$	9–7	69.4	54.3	55.5	114.8	85.5	39.4	62.6
Snap Corn	10–7	72.4	76.0	87.0	140.5	92.3	65.5	76.5
Av. of silages preserved with:								
Snap Corn		83.1	85.0	92.0	178.1	94.5	76.1	85.4
$Na_2S_2O_5$		79.8	72.2	79.4	113.4	91.5	58.1	103.3

[a] Second number indicates year in which silage was made.

general influence of the two preservatives was quite pronounced. When Silages 4–6, 6–7, and 9–7 are averaged and compared with the mean of Silages 5–6, 7–7, and 10–7, the ground snap corn preserved 13% more dry matter than the sodium metabisulfite. This additional preservation was represented in all fractions except ash, and was most evident in ether extract and N.F.E., indicating a better preservation of the carbohydrates.

Such a preservation may have a marked influence on the usefulness of a preservative. To determine if this difference in preservatives is of real significance, a comparison of the efficiency of dry matter preservation is presented (Table 4). Using the data for the silage preserved with sodium metabisulfite as a basis for comparison, the table indicates that, on the average, for each 15 lb.

TABLE 4

Stored losses of forage and additives

	Material	Dry matter	Storage loss	Recovered dry matter
		(lb.)		
Forage stored with $Na_2S_2O_5$ and snap corn stored in a bin:				
Forage	2,000	460	138	322
Snap Corn	200	180	0	180
Forage stored with added snap corn:				
Forage	2,000	460	69	391
Snap Corn	200	180	15	165
Net gain of treatment:				
Forage dry matter	+69			
Snap Corn	−15			

of snap corn lost during silage fermentation there was a net gain of 69 lb. of forage dry matter in the silos preserved with ground snap corn. This increase in preservation may or may not be of economic importance, depending upon the relative value of ground snap corn and silage dry matter. The comparison does not necessarily reflect the true differences, because it does not evaluate the relative feeding value of the two resulting rations.

Feeding value of the silages. The objectives of the feeding trials were (1) to determine which of the silages was most suitable for dairy cows and (2) to create differences in response wide enough to differentiate between stages of maturity and preservatives. This dual function of the feeding trial created problems which were not entirely overcome in this experiment. In attempting to feed against obvious protein deficiencies due to stage of maturity, actual differences may have been narrowed. The feeding trial data are also complicated by differences in total dry matter intake which resulted from free-choice feeding of the silage. Earlier feeding trials using oat silage had indicated the silage to be a low-energy feed; for this reason, no change in the grain fed was made with the ground snap corn silages. Once started, this feeding plan was continued throughout the trials. Intake and production data are shown (Table 5).

In general, there was no consistent difference in silage intake due to preservative. The difference in silage dry matter resulted in a mean difference in dry matter intake of 4 lb. per cow per day in favor of the snap corn. In general, silage consumption increased with increasing stage of maturity. This increase continued through the milk stage in 1956 and through the prebloom stage in 1957. The consumption and utilization of the milk-stage oats illustrate the variation which may be encountered between years. Silage 3–7 was ensiled in 1957, which was a very poor year for oats. The crop had suffered heavy disease damage and, by the time it was in the milk stage, heavy leaf damage had occurred.

TABLE 5

Rations fed and animal response to the silages

Stage of maturity and preservative	Silage No.	Av. daily intake per cow				Silage per 1,000 lb. live weight	Beginning milk production	28-day change	
		Silage	Hay	Grain	Total dry matter			FCM	Body weight
		(lb)						(%)	(lb.)
Dough									
$Na_2S_2O_5$	1–6	50.4	0	5.4	26.0[b]	50	26.2	−10	−12
Milk									
$Na_2S_2O_5$	2–6	89.4	0	5.0	27.5	88	26.3	−12	+10
None	3–7	52.0	0	6.1	20.4	59	24.3	−30	−16
Prebloom									
$Na_2S_2O_5$	4–6	75.6	8.0	9.0[a]	34.0	72	27.6	−22	− 8
Snap Corn	5–6	82.3	8.0	7.8[a]	37.9	77	23.1	− 6	+10
$Na_2S_2O_5$	6–7	84.5	0	4.7	25.3	98	27.8	−26	+23
Snap Corn	7–7	80.6	0	4.7	26.8	96	28.3	− 9	+35
None	8–7	83.1	0	7.0	27.1	87	27.5	− 8	−15
Boot									
$Na_2S_2O_5$	9–7	66.4	0	4.3	16.5	71	25.9	−10	− 1
Snap Corn	10–7	66.7	0	4.3	22.5	71	25.6	− 9	+ 1

[a] Includes 2 lb. cottonseed meal.
[b] Two hours of grazing on winter oats.
[c] Second number indicates year in which silage was made.

The estimated grain yield would have been cut by at least 50%. The prebloom oats showed much less variation and this factor may be of considerable importance in deciding the stage to cut oats, particularly in a year such as 1957. The cows consumed sufficient nutrients on all silages to meet accepted feeding standards, but animal response would seem to indicate that factors other than TDN and digestible protein were involved.

Animal response as measured by milk production was satisfactory on all silages preserved with ground snap corn, but only the oats ensiled at the boot stage resulted in satisfactory milk production when preserved with sodium metabisulfite. The one comparison with oats ensiled without preservative (Silage 8–7) indicated that sodium metabisulfite resulted in a less valuable silage than a well-made silage without preservative. This comparison brings out a factor of considerable importance to silage experiments, because the value of a preservative depends upon the value of the control silage. Had the silage without preservative been poorly fermented, the comparison between the silages could have been quite different.

Stage of maturity had little influence on milk production, so long as the forage was cut prior to the milk stage. The milk-stage silage failed to support a normal rate of milk production both years. The one silage cut at the dough stage was unable to support normal milk production, although fed with high-quality pasture.

When both milk production and preservation of nutrients were considered, the optimum stage for cutting oats was in the prebloom stage.

CONCLUSIONS

Ten silos were used to compare stage of maturity and two preservatives for oat silage. The following conclusions are valid: Optimum preservation of nutrients and feeding value was obtained at the prebloom stage of maturity. The use of 200 lb. per ton of ground snap corn improved nutrient preservation and resulted in silage of acceptable feeding value. Sodium metabisulfite was an unsatisfactory preservative for oat silage. The value of this preservative under conditions of poor silage-making was not studied.

ACKNOWLEDGMENTS

The authors wish to express their appreciation to Margaret Borders for aid in the laboratory, to Harold Childres for supervising the feeding trials, and to H. S. Johnson, Jr., for analysis of the silages.

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BORON IN COWS' MILK[1]

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SUMMARY

Boron appears to be a natural constituent of cows' milk, the amount varying with the intake of the element in the feed. Three hundred milligrams of boron fed daily as sodium tetraborate for a period of 2 mo. to six cows increased the average amount of boron in the milk approximately 2.5 times, from 270 to 663 μg. per liter of milk. The total daily amount of boron thus secreted appeared to be rather constant for individual cows, regardless of the amount of milk produced. A consistent increase in boron content of the milk was noted from the 1st to the 2nd mo. in each half of the trial, regardless of breed, group, or treatment. No plausible explanation for this increase fits the situation as a whole. A study of partition of the boron between cream and skimmilk indicated that about one-fourth of the total amount was in the cream and three-fourths in the skimmilk.

Boron is an essential element for plants and is added to fertilizers for specific crops in certain areas, especially where agriculture is highly intensified. Alfalfa and sainfoin *(10)*, soybean meal, and beet pulp *(7)*, in comparison with some common grasses *(6, 9)*, contain considerable amounts of boron. Previous investigators *(1)* have shown that boron is widely distributed in animal tissues.

Several earlier investigators *(2, 3, 4, 13)* detected its presence spectrographically in the ash of milk, but did not determine the amount. More recently Hove *et al. (8)* reported values ranging from 94 to 228 μg. per liter of milk; they concluded that the amount is influenced by the boron content of the feed. Owen *(12)* found that the addition of 20 g. of borax to the daily ration of milk cows increased the boron content of the milk from 0.7 to over 3.0 p.p.m.

EXPERIMENTAL PROCEDURE

This work was done during the winter of 1956–57 with six cows divided into two groups of three each, representing the Ayrshire, Guernsey, and Holstein breeds. Each breed pair was matched as closely as possible with respect to stage of lactation. The supplement fed was borax ($Na_2B_4O_7 \cdot 10\ H_2O$) in a daily amount of 2.6 g., equivalent to 300 mg. of elemental boron. One group received the supplement during December and January, the other during February and March.

Composite two-day milk samples of one liter were taken from each cow about the middle of each month. Triplicate 300-ml. aliquots were placed in silica dishes, mixed with a slurry of 1 g. CaO and H_2O, and evaporated to dryness on a steam bath. The samples were then ashed in an electric furnace at approximately 600° C. and the ash dissolved in 6 *N* H_2SO_4. Boron was determined photometrically by the method of Oelschläger *(11)*. Reproducibility of the method was

Received for publication January 3, 1958.

[1] Contribution No. 1140.

determined by adding 100 and 200 μg. of boron to 300-ml. aliquots of normal herd milk. Percentage recoveries were 90, 95, 90, and 105; average, 95%.

RESULTS

Results are summarized (Table 1). The boron content when the cows were on the control ration averaged 270 μg. per liter; the range was from 193 to 360 μg/liter. When the supplement was being fed, the average value was 663 μg/liter, with a range from 510 to 860 μg/liter. The increase was highly significant. One cow (A105) did not show any increase when fed the supplement. No reason can be assigned for this; therefore, the values for this cow, for February and March, are not included in the average. The average level of boron in the milk when the supplement was fed was approximately 2.4 times that found when the cows were on the control ration.

TABLE 1

Effect of feeding cows sodium tetraborate on boron content of milk

	Cows on control ration				Cows receiving supplemental boron			
	First half of season							
	A105	G104[a]	H71	Average	A71	G100	H70[b]	Average
	(μg. boron/liter milk)							
December	260	267	243	261	677	593	510	593
January	360	.....	293	327	860	660	527	682
Average—first half	310	.....	268	285	769	627	519	638
	Second half of season							
	A71	G100	H39	Average	A105[c]	G86	H71	Average
	(μg. boron/liter milk)							
February	260	260	193	238	243	743	593	668
March	268	327	243	279	293	777	693	735
Average—second half	264	294	218	259	268	760	643	702
Average—entire season	287	281	241	270 ±13[d]	.....	693	581	663 ±35[d]

The initial letter prefixed to each cow's number indicates the breed.

[a] Through a misunderstanding, this cow was slaughtered before the January samples were taken; Cow G86 was substituted.

[b] Slaughtered because of repeated attacks of acute mastitis; Cow H39 was substituted in February.

[c] Not included in the average.

[d] Standard error of the mean.

The results from the control cows are of the same order of magnitude as those obtained by Falckenthal *(6)*; they are somewhat higher than Hove's values *(8)* (94–228 μg. per liter), possibly owing to the fact that considerable beet pulp was fed. This material has been shown *(7)* to be relatively high in boron.

The results when the supplement was fed are somewhat lower than those reported by Owen *(12)*, especially with respect to maximum value (860 μg. per liter in contrast to 3,000). However, Owen fed 20 g. of borax daily, or nearly

eight times as much as was fed in this investigation, confirming the conclusion of Hove *et al. (8)* that the amount of boron in milk is influenced by the amount in the feed.

The daily output of boron in the milk, based on the milk yield during the two-day sampling periods, is shown (Table 2). Individual cows, whether or not they were receiving the supplement, showed a rather constant output of boron in their milk.

TABLE 2

Boron output in the milk

(Based on average two-day milk yield for the several sampling periods)

	Cows on control ration			Cows on boron-supplemented ration		
	First half of season					
	A105	G104	H71	A71	G100	H70[b]
	(μg. boron daily)					
December	4.37	2.75	5.53	12.56	6.80	8.79
January	5.22	([a])	5.34	12.73	6.92	7.50
	Second half of season					
	A71	G100	H39	A105[c]	G86	H71
	(μg. boron daily)					
February	3.39	2.98	3.74	3.36	11.85	10.29
March	3.64	3.31	4.51	3.50	12.11	11.25

[a] See footnote [a] under Table 1.
[b] See footnote [b] under Table 1.
[c] Low values were due to the low level of boron in this cow's milk.

It is of interest also to note that without exception, and regardless of whether or not the cows were receiving the boron supplement, the values in each cow's milk for the second month of each half of the trial were somewhat higher than those for the first month. These increases ranged from 8 to 183 μg/liter, or from 3.1 to 38.5% ; most of the increase can be explained on the basis of the results in Table 2 *(q.v.)*. The rather constant output of boron that accompanied the normal decrease in milk yield accounts largely for these higher values in the second month.

Partition of the boron between fat and solids-not-fat of the milk was also investigated. It was found that the boron content of cream (40% fat) was about 3.6 times higher than that of the skimmilk. On this basis, ordinary fluid milk containing 4% fat would have about one-quarter of the total amount of boron in the cream, the remaining three-quarters being in the skimmilk.

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EFFECT OF POTASSIUM NITRATE INTAKE ON DAIRY CALVES[1]

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SUMMARY

This study provides information on the effects of level, frequency, and duration of nitrate intake on the nitrate tolerance of young dairy animals. Administration of KNO_3 to bull calves indicated that the minimum lethal dose for initial intake was in excess of 30 g. per 100 lb. body weight. Toxic response to the compound was found to vary between animals, but was not materially affected by experimental method of intake (by capsule or dissolved in milk). The level of KNO_3 ingestion exerted little, if any, influence on the time required for the in vivo formation of maximal levels of methemoglobin (determined spectrophotometrically). Up to 1.3 g. % nitrate, but no nitrite, was detected by microbiological assay of the urine from two calves receiving 25 g. KNO_3 per 100 lb. body weight and showing low levels of methemoglobin.

During recent years, nitrate poisoning has become increasingly important to the livestock industry, either by virtue of an increased incidence or owing to more accurate diagnosis of the symptoms. Most of the early work indicates that this condition was associated primarily with the ingestion of oat hay or corn stalks high in KNO_3 equivalent *(2, 5, 9, 15–17)*. More recently, other feed materials such as corn silage, sorgo silage, and alfalfa hay also have been implicated in nitrate poisoning of cattle and sheep, when the KNO_3 equivalent on a dry matter basis exceeded approximately 0.7% of the feedstuff.

Davidson *et al. (5)* reported that the symptoms of nitrate poisoning were quite uniform in character, and that the severity of these symptoms was markedly increased if the animals were disturbed, or forced to move about. The symptoms included an increased pulse rate, weakness, staggering gait, apparent blindness in some cases, and cyanosis; in severe cases, death usually occurred with little or no struggling. Similar symptoms were reported by Muhrer *et al. (13)*, with the exception that motor spasms progressed rapidly to convulsions, terminating in death. However, in the latter report, nitrate was only one of a group of many factors believed to be responsible for the extreme toxicity of forages produced in a drought area. Beath *et al. (1)* have reported similar symptoms.

Ingested nitrates are believed to be converted to nitrites in the digestive tract *(1, 12)*, and the nitrites then absorbed by the blood stream cause the hemoglobin to be converted into methemoglobin, which is incapable of carrying oxygen to the tissues *(1, 6, 12, 13, 14)*. The methemoglobin will gradually be reduced to normal hemoglobin in drawn blood samples *(6, 10)*.

Relatively little information is available on the importance of nitrate poisoning in young animals and on its relationship to their later growth and development. However, Bradly *et al. (4)* report that the minimum lethal dose of KNO_3

Received for publication August 28, 1957.

[1] Contribution from the Missouri Agricultural Experiment Station, Journal Series No. 1787. Approved by the Director.

(by stomach tube) for dairy calves is approximately 25 g. per 100 lb. body weight, with large quantities of nitrite being found in the bile and urine of animals receiving the nitrates.

METHODS

Bull calves from the University of Missouri dairy herd were used as experimental animals. The calves nursed their dams for the first three days after birth and were bucket-fed thereafter, receiving whole milk (10% of body weight daily) to the age of 1 mo., followed by a gradual change to skimmilk at a daily rate of 10% of body weight, until a maximum of 14 lb. skimmilk was reached. Grain was fed free choice, with the older calves consuming approximately 4 lb. per day. Alfalfa hay, containing less than 0.21% KNO_3 equivalent on a dry matter basis, was fed ad libitum.

Potassium nitrate dosages were administered either by gelatin capsule or by dissolving the compound in skimmilk.

Blood samples were drawn from the jugular vein into sterile tubes containing 1.5 mg. potassium oxalate per milliliter of blood.

Analysis for hemoglobin and methemoglobin was started within 30 min. after the sample was drawn, since methemoglobin in stored blood is converted back slowly to functional hemoglobin *(10)*. A Coleman Model 14 Spectrophotometer was used with the method of Evelyn and Malloy *(7)*, for determining hemoglobin and methemoglobin.

Urine samples were collected from a limited number of calves, using a modification of the urine-collection apparatus as designed by Hobbs *et al. (11)*. Urine samples were held in a frozen state (−15° C.) between collection and analysis for nitrate and nitrite content. The microbiological method developed by Garner *et al. (8)* was used in determining the nitrate and nitrite content of the alfalfa hay and urine samples.

OBSERVATIONS AND RESULTS

Individual animals vary markedly in the time that maximum conversion of hemoglobin to methemoglobin occurs following nitrate ingestion (Table 1). However, neither the level of nitrate intake nor the mode of intake (gelatin capsules vs. milk) appears to have much influence on the time of maximum conversion. The slight increase in methemoglobin formation with KNO_3 given in capsules may be, in part, owing to a carryover in toxic effect from the dosage given in milk during the preceding week. Most of the calves refused to drink milk containing more than 1.9 g. % KNO_3.

In the experimental trials, maximum conversion occurred approximately 5 hr. after nitrate intake. In contrast to the 25-g. minimum lethal dose reported by Bradly *et al. (4)*, no calves died in this series of feeding trials, even though doses up to 30 g. per 100 lb. body weight were given. A possible explanation for the difference in tolerance could be owing in part to the level of energy in the ration and to the condition of the calves at the time of feeding potassium nitrate.

The response (conversion of hemoglobin to methemoglobin) of calves to initial KNO_3 intake shows considerable individual variation, but also exhibits a general trend of increase in toxic symptoms with increasing KNO_3 dosage levels (Table 1). The maximum methemoglobin values at 10, 15, 20, and 30 g. per 100 lb. body weight dosages were 0, 8, 16, and 37%, respectively.

In all cases where animals received nitrates, and showed methemoglobin formation, there was an increase in heart rate (Figure 1).

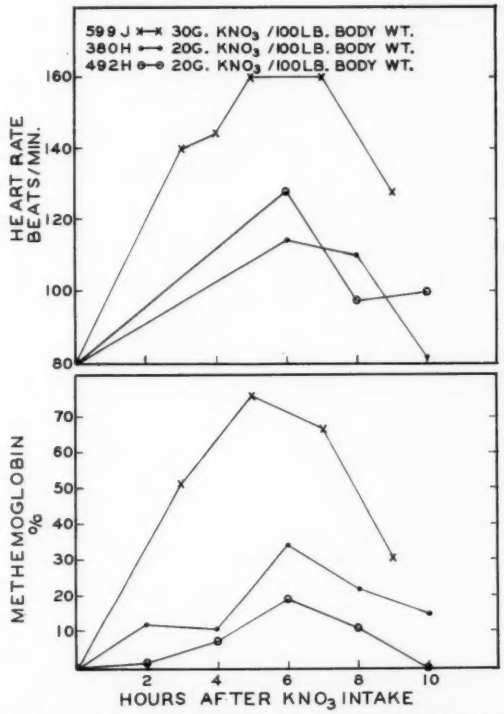

FIG. 1. Relationship between heart rate and methemoglobin formation.

Large quantities of nitrates were found to be excreted in the urine of calves receiving 25 g. KNO_3 per 100 lb. body weight (Figure 2). However, no nitrites were detected in the urine at this level of KNO_3 intake. As shown in Figure 2, excretion of nitrates in the urine, and conversion of hemoglobin to methemoglobin in blood, occur at approximately the same time following KNO_3 ingestion.

Two calves were given multiple doses of potassium nitrate (15 g. per 100 lb. body weight) over a period of eight days. The calf receiving a single daily KNO_3 dose gained an average of 2 lb. in body weight per day and showed less than 10% maximum conversion of hemoglobin to methemoglobin on any given day. The other calf, receiving a total of 30 g. per 100 lb. body weight daily, in two equal portions given 12 hr. apart, neither gained nor lost weight during the

TABLE 1

Time of maximum hemoglobin-to-methemoglobin conversion following potassium nitrate intake

Calf No.	Age	Body wt.	Dosage level	Method of KNO_3 intake	Maximum conversion HB to MHB	Hr. after intake when maximum conversion occurred
	(wk.)	*(lb.)*	*(g. KNO_3/cwt)*		*(%)*	
492H	6	155	10	Capsule	0	....
	9	200	20	Capsule	19	6
380H	5	147	10	Capsule	0	....
	8	190	20	Capsule	34	6
490H	8	188	15	Capsule	8	5
460H	7	168	15	Capsule	4	5
498H	8	146	20	Milk	0	....
	9	162	20	Capsule	0	....
	10	164	25	Milk	5	4
	11	181	25	Capsule	19	8
495H	9	158	20	Capsule	5	5
	10	174	20	Milk	1	7
	11	185	25	Milk	3	4
	12	200	25	Capsule	4	4, 6
485H	10	181	20	Capsule	16	3
	12	206	25	Milk	4	4
	13	223	25	Capsule	11	4
599J	15	172	30	Capsule	37	9
	18	198	30	Capsule	76	5
	20	224	25	Milk	3	4, 6, 8
	21	238	25	Capsule	3	4

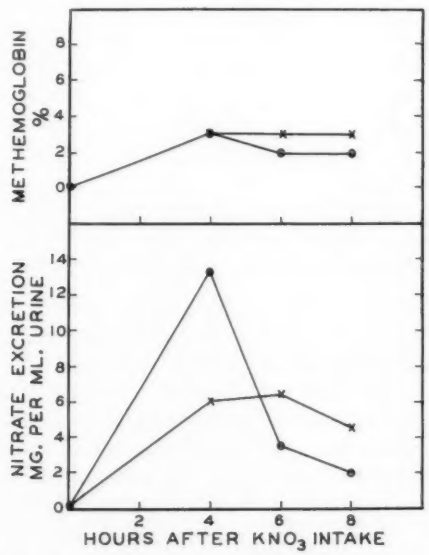

FIG. 2. Urinary nitrate excretion and methemoglobin formation in two calves receiving 25 g. KNO_3/100 lb. body weight.

eight-day experiment. The latter calf showed maximum hemoglobin-to-methemoglobin conversion values of between 35 and 50% on four of the eight days.

The toxic symptoms observed throughout this study, for both the single and multiple KNO_3 doses, were relatively mild, as compared to nitrate poisoning symptoms generally seen both in dairy and in beef cattle under practical farm conditions, at much lower levels of nitrate intake. This immediately poses a question as to the true nature of so-called nitrate poisoning observed on the farm. The apparent discrepancy between symptoms induced experimentally and those observed under practical conditions may be owing either entirely or in part to: *(1)* Form of nitrate ingested (inorganic vs. organic); *(2)* diet, rumen function, and nutritional status of animals in conjunction with amount of environmental stress; *(3)* age and factors associated with stress of milk and meat production, and *(4)* level of nitrate intake, particularly as related to total period of intake and amount consumed at any given time.

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COMPARATIVE METABOLISM OF CALCIUM AND STRONTIUM IN LACTATION

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SUMMARY

The comparative metabolism of calcium and strontium in the lactating and nonlactating goat was studied, using double tracer methods. At steady state, calcium was preferentially secreted into milk by a factor of about 11 over strontium in its passage from diet to milk. With corollary data from nonlactating animals, it was found that the largest amount of discrimination took place in the gastrointestinal absorption of these alkaline earths, calcium being absorbed from three to four times that of strontium. The over-all discrimination of 0.09 between milk and diet resulted primarily from the following discrimination factors: absorption, 0.24; urinary excretion, 0.60; transfer from plasma to milk, 0.61. Under steady state conditions, the relative Sr°/Ca° ratios were diet = 1, plasma = 0.23, skeleton = 0.23, milk = 0.09, and urine = 4.0. These values indicate that there is no differential movement of calcium and strontium between blood and bone, and that strontium is preferentially excreted in the urinary process.

Radiostrontium is at present detectable by low-level counting techniques in the food chain (soil, plants, milk, animals, and man); this radioactivity originates from nuclear detonation *(8)*. The progression of radiostrontium along the food chain generally follows, and is related to calcium movement *(3, 12)*. The amount of radiostrontium in the human population for a given amount of fallout will depend on many factors, especially the differential movement of calcium and strontium in the food chain and the strontium^{90} content of the dietary calcium. In the United States, it has been estimated that from 70 to 80% of the calcium in the human dietary originates from milk or milk products. Fortunately, considerable discrimination against strontium in favor of calcium occurs in the passage of these alkaline earths from the diet of the dairy animal to the milk. Were it not for this differential metabolism, the Sr^{90} content of the human population would be about five times higher than otherwise.

It has been shown that the Sr^{90}-to-Ca ratio of milk will be from 0.09 to 0.16 that of the ration being consumed by the animal. This was determined by experimental studies using radioisotopes of strontium and calcium *(4)*, analysis of feed and milk for fallout Sr^{90} and stable calcium *(1)*, and analysis of feed and milk for stable calcium and stable strontium *(6)*. The present paper presents confirmation of the over-all discrimination in the goat; in addition, more detailed studies are described that give information on the comparative metabolism of these two elements in a ruminant, with emphasis on the physiological mechanisms responsible for their differential behavior.

Received for publication February 7, 1958.

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MATERIALS AND METHODS

The terminology used in discussing the comparative movement and differential handling of calcium and strontium is that proposed by Comar *et al. (5)*. The "Strontium-Calcium Observed Ratio" is defined as the ratio of strontium to calcium in any product compartment, divided by the ratio in a precursor compartment, as follows:

$$OR_{\text{sample-precursor}} = \frac{\text{Sr/Ca of sample}}{\text{Sr/Ca of precursor}} \qquad (1)$$

The concentrations of Ca* and Sr* in tissues or excretions are expressed as per cent of administered dose.

Lactating goats—daily isotope ingestion. Two lactating goats, each of about 36 kg. body weight, were placed in metabolism stalls, as described by Comar *(2)*, for the quantitative collection of feces and urine. A good-quality mixed hay was fed with a commercial dairy feed containing 16% crude protein. After a preliminary conditioning period of about a week, the simultaneous oral administration of Ca^{45} and Sr^{89} by capsule was begun. Each goat received 200 μc. Ca^{45} and 66 μc. Sr^{89} twice daily by capsule, at 8 A.M. and 4 P.M. for 12 days;

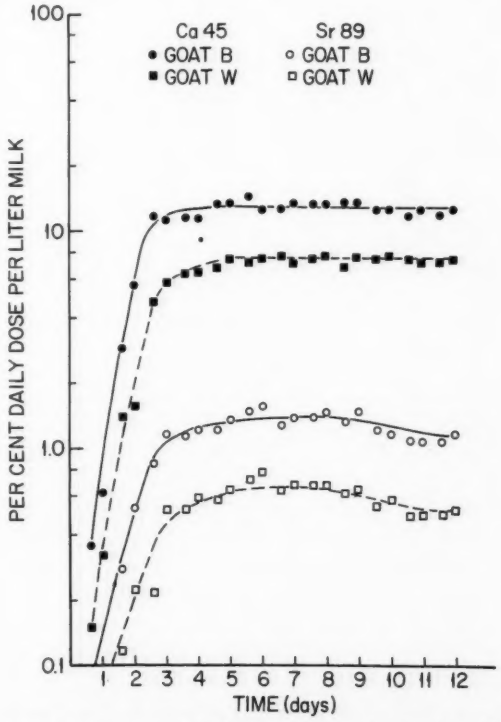

FIG. 1. Levels of daily ingested Ca^{45} and Sr^{89} appearing in the milk of goats.

the isotope preparations were essentially carrier-free and in the form of the chloride salt. The feces and urine were collected only once daily, and the animals were milked twice daily. On the 13th day, the goats were killed and tissue samples were obtained. The milk, tissues, and excretions were analyzed for Ca^{45} and Sr^{89} by the differential absorption procedure *(2)* and results expressed as "per cent of the daily ingested dose."

Nonlactating goats—single-dose studies. As will be pointed out later, it was necessary to compare results after oral and intravenous administration, to obtain information on the role of gastrointestinal absorption in causing discrimination. Single oral doses of Ca^{45} and Sr^{89} were given by gavage to two goats; heparinized blood samples were taken from the jugular vein at periodic intervals over a three-day period. Two other nonlactating goats of comparable body size were given 600 μc. Ca^{45} and 60 μc. Sr^{85} intravenously. Frequent blood samples also were obtained from these animals through an indwelling catheter placed in the jugular vein. The time intervals of blood sampling from both methods of administration were as shown (Fig. 2). The plasma of the "oral" group was assayed for the two beta-emitting isotopes, Ca^{45} and Sr^{89}, by the differential absorption method. In the intravenous group, Ca^{45}, a pure beta emitter, was determined by conventional beta-counting methods with a Geiger-

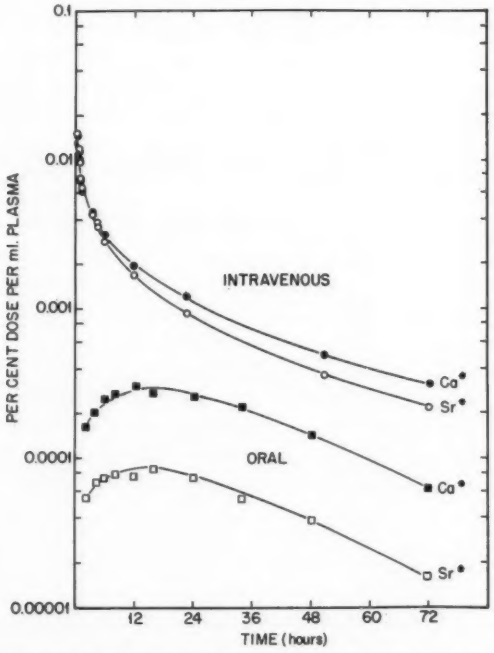

FIG. 2. Levels of Ca* and Sr* in the plasma of goats after a single oral or a single intravenous dose of the radioisotopes.

Muller detector and Sr^{85}, a pure gamma emitter, by liquid counting in a well-type scintillation detector. At the levels of activity used, the beta count did not contribute to the gamma count, and vice versa. At five days after dosage, the intravenously injected goats were killed and various bone samples were selected for radioassay.

RESULTS AND DISCUSSION

Lactation. As shown (Figure 1), the Ca^{45} and Sr^{89} content of the milk, expressed as per cent of the daily dose per liter, increased rapidly until a plateau level was attained at about three days. After this time, no appreciable change in the milk levels occurred. A slight decrease in the Sr^{89} levels between the 9th and the 12th day was observed; if real, no physiological explanation can be offered. It should be pointed out also that the Ca^{45} and Sr^{89} concentrations in the plasma did not change after three to four days of daily radioisotope administration, and that the daily excretion of these isotopes in the urine and feces likewise remained essentially the same. Thus, these animals may be considered at "steady state", in that the radioisotope concentrations in the body fluids and excretions had attained some constant function of the intake. Under these conditions, the experimental animals were secreting about 10% of the Ca^{45} and about 1% of the Sr^{89} ingested per day per liter of milk (Figure 1 and Table 1).

TABLE 1

Summary of levels of Ca^{45} and Sr^{89} in plasma and milk and observed ratios in the lactating goat at steady state

	Goat B	Goat W
Ca^{45} in milk (% of daily dose/liter)	12.8	7.3
Sr^{89} in milk (% of daily dose/liter)	1.3	0.61
Ca^{45} in plasma (% of daily dose/liter)	0.87	0.58
Sr^{89} in plasma (% of daily dose/liter)	0.24	0.12
$OR_{milk\text{-}diet}$[a]	0.10	0.083
$OR_{plasma\text{-}diet}$	0.27	0.20
$OR_{milk\text{-}plasma}$	0.38	0.41

[a] $OR_{sample\text{-}precursor} = \dfrac{Sr^*/Ca^* \text{ in sample}}{Sr^*/Ca^* \text{ in precursor}}$.

Under steady-state conditions, a comparison of the various OR values (calculated from Equation 1) indicates the degree of discrimination occurring in the movement of Ca^{45} and Sr^{89} among compartments (diet, plasma, and milk). These have been summarized (Table 1). Of considerable importance is the overall differential movement of calcium and strontium from diet to milk. The $OR_{milk\text{-}diet}$ was found to be 0.10 and 0.08 for Goat B and Goat W, respectively, indicating that dietary calcium was preferentially secreted into milk by a factor of ten to 12 times that of dietary strontium. These values are in agreement with the $OR_{milk\text{-}diet}$ values previously reported for the bovine, which averaged about 0.13. In the movement from plasma to milk, calcium was preferentially secreted by a factor of 2.5 over strontium as shown by the $OR_{milk\text{-}plasma}$ values of 0.38 and 0.41. In the cow, the $OR_{milk\text{-}diet}$ was found to be 0.14, the $OR_{plasma\text{-}diet}$, 0.37, and the $OR_{milk\text{-}plasma}$, 0.38 *(3)*; from these data, it is apparent that the

differential movement of calcium and strontium from plasma to milk in the cow was identical with that in the goat.

From a knowledge of the specific activities of Ca^{45} in the diet and the milk of the goats, it was possible to estimate the comparative contribution of dietary calcium and skeletal calcium to milk calcium by applying the usual principles of isotope dilution. The specific activities of the diet and milk averaged 8.0 and 6.8% of the daily ingested dose of Ca^{45} per gram of total calcium, respectively; from these values, it was calculated that approximately 85% of the milk calcium originated from the diet and 15% primarily from the maternal skeleton. Thus, milk calcium is comprised essentially of dietary calcium; this same relationship would hold for radiostrontium, since there are negligible differences in the movement of calcium and strontium between blood and bone *(5, 10, 14, 16)*. From these considerations, it becomes evident that, after a few days of daily ingestion, the milk levels of Sr^{90} represent mainly the activity that was ingested over the previous few days and would constitute an immediate and contemporary indication of the fallout Sr^{90} available to the lactating animal. This is also borne out by the fluctuations observed in the fallout Sr^{90}-to-calcium ratios in fresh milk samples taken from the Chicago dairies; these changes were correlated with nuclear tests and their relation to seasonal variations in feeding practices *(15)*.

Absorption and excretion. The levels of Ca^{45} and Sr^{89} in per cent of daily dose per day, in the milk, excretions, and body of the goat at steady state, are summarized (Table 2). The fecal excretions of both calcium and strontium were high, as judged from direct measurement—87.5 and 101% of the ingested dose,

TABLE 2

Levels of Ca^{45} and Sr^{89} in milk, excretions, and body of the goat at steady state (in per cent of daily dose per day) and observed ratios

	Goat B	Goat W	Mean
Milk			
Ca^{45}	14.0	6.8	10.4
Sr^{89}	1.4	0.57	1.0
Feces (direct measurement)			
Ca^{45}	85	90	87.5
Sr^{89}	94	108	101
Feces (by difference[a])			
Ca^{45}	77	82	79
Sr^{89}	94	95	95
$OR_{feces\text{-}diet}$[b]	1.13	1.12	1.12
Urine			
Ca^{45}	0.42	0.74	0.58
Sr^{89}	2.3	1.8	2.1
$OR_{urine\text{-}diet}$	5.5	2.5	4.0
$OR_{urine\text{-}plasma}$	20.5	12.1	16.3
In body (estimated from bone analysis)			
Ca^{45}	9.1	11	9.8
Sr^{89}	2.1	2.2	2.2

[a] Intake—(estimated amount in body + urinary excretion + milk secretion).

[b] $OR_{sample\text{-}precursor} = \dfrac{Sr^*/Ca^* \text{ in sample}}{Sr^*/Ca^* \text{ in precursor}}$.

respectively. Because of the unrealistic value of 108% obtained for Goat W, estimates were made by calculating the amounts retained in the body from bone analyses and subtracting the sum of this value, the urinary excretion, and the milk secretion from the intake. These values (feces, by differences) appeared to be quite reasonable. Because of the high fecal excretions and the inherent inaccuracies, an alternate experiment was done to give estimates of differential absorption. In this study, a comparison was made of plasma levels of radiostrontium and radiocalcium, after oral and intravenous administration to nonlactating goats. The data are given (Figure 2).

Comparison of the plasma levels (Figure 2) shows clearly that there was little difference between radiocalcium and radiostrontium concentrations following injection; after oral administration, however, the plasma levels of radiocalcium were significantly higher than those of radiostrontium. It is apparent, then, that radiostrontium was being absorbed less effectively than was radiocalcium. The extent of the absorptive discrimination was estimated by comparing the $OR_{plasma\text{-}dose}$ values after both modes of administration. The $OR_{plasma\text{-}dose}$ after intravenous injection was 1.0 for the first 6 hr.; thereafter, it decreased until a value of 0.7 was attained at 72 hr. In the goats receiving the radioisotopes by the oral route, the $OR_{plasma\text{-}dose}$ was about 0.3 when absorption was maximum at about 12 hr., and decreased to a value of about 0.27 at 72 hr. Thus, it is inferred that the nonlactating goat was able preferentially to absorb radiocalcium over radiostrontium by a factor of about three, and that this also would be true for the lactating animal.

The preferential absorption of radiocalcium over radiostrontium by the goat is contrary to reports of equal absorption of these cations in the cow *(11)*. However, the data in the cow were obtained from single-dosage studies of one isotope at a time to the same animal on different occasions or to different animals. Therefore, they are not so precise as double-tracer studies. Further investigations are needed to answer the question of gastrointestinal discrimination in the bovine.

In the urinary process (as shown in Table 2), strontium was preferentially excreted as shown by the $OR_{urine\text{-}diet}$ values of 5.5 and 2.5; the comparative plasma clearance by the kidney was 12 to 20 times greater for strontium than for calcium. These comparative clearance values differ from those of three to five found in man and rats, but agree with the clearance figure of about 24 obtained in cattle *(4)*.

Tissue analysis. The strontium-calcium observed ratios, and the Ca^{45} and Sr^{89} content of various tissues from the lactating animals, are shown (Table 3). First, it may be noted that the $OR_{tissue\text{-}diet}$ values for the bone samples were relatively uniform, although considerable variation was found in the per cent of the daily ingested activities present. In Goat B, for example, the Ca^{45} and Sr^{89} contents of the jawbone were almost three times those of the femur shaft; yet, the $OR_{tissue\text{-}diet}$ values for both bones were almost identical. The teeth, on the other hand, had an observed ratio higher than that of the other bony tissues, probably caused by the exchange of Ca^{45} and Sr^{89} directly into the tooth enamel

TABLE 3

Levels of Ca^{45} and Sr^{89} and observed ratios (OR) in various tissues of the lactating goat at steady state

Tissue	Ca^{45} (% daily dose per g. tissue)	Sr^{89} (% daily dose per g. tissue)	$OR_{tissue\text{-}diet}$[a]
	Goat B		
Femur shaft	0.014	0.0035	0.26
Femur epiphysis	0.018	0.0035	0.20
Rib shaft	0.017	0.0045	0.26
Rib epiphysis	0.033	0.0070	0.21
Jaw (angle)	0.037	0.0092	0.25
Incisor tooth	0.010	0.0037	0.36
Plasma	0.00087[b]	0.00024[b]	0.27
	Goat W		
Femur shaft	0.022	0.0045	0.20
Femur epiphysis	0.021	0.0041	0.20
Rib shaft	0.025	0.0057	0.22
Rib epiphysis	0.025	0.0054	0.22
Jaw (angle)	0.055	0.0107	0.20
Incisor tooth	0.0056	0.0018	0.32
Plasma	0.00058[b]	0.00012[b]	0.20

[a] $OR_{tissue\text{-}diet} = \dfrac{\text{Sr/Ca tissue}}{\text{Sr/Ca diet}}$.

[b] Plasma values were given as % daily dose per ml.

from the bolus during rumination; the $OR_{bolus\text{-}diet}$ should be close to unity. It should also be noted that the $OR_{plasma\text{-}diet}$ and $OR_{skeleton\text{-}diet}$ values were quite similar in both animals; the average $OR_{skeleton\text{-}plasma}$ value of 0.95 (Table 2) demonstrates that no appreciable discrimination occurred in the blood-bone system. This has been previously shown in rats *(5, 16)*, rabbits *(14)*, and embryonic-chick bone cultures *(10)*.

The Ca^{45} and Sr^{85} analyses of bone samples from the singly-injected, nonlactating goats are presented (Table 4). These data again demonstrate the variability with which different types of bones accumulate radiocalcium and

TABLE 4

Levels of Ca^{45} and Sr^{85} and observed ratios (OR) in the skeleton of the nonlactating goat after intravenous administration

Tissue	Ca^{45} (% dose/g fresh wt.)	Sr^{85} (% dose/g fresh wt.)	$OR_{tissue\text{-}dose}$[a]
	Goat 1		
Tibia epiphysis	0.0071	0.0055	0.77
Tibia shaft	0.0056	0.0041	0.73
Rib epiphysis	0.020	0.039	0.52
Rib shaft	0.014	0.021	0.64
	Goat 2		
Tibia epiphysis	0.0078	0.0064	0.83
Tibia shaft	0.0090	0.0074	0.83
Rib epiphysis	0.044	0.033	0.76
Rib shaft	0.027	0.019	0.71
Incisor tooth	0.0045	0.0038	0.83

[a] $OR_{tissue\text{-}dose} = \dfrac{\text{Sr/Ca of tissue}}{\text{Sr/Ca of dose}}$.

radiostrontium; also, the similarity in the strontium-calcium observed ratios. This can be illustrated by reference to the amounts of activity present in the tibia shaft and rib shaft of Goat 1; the latter tissue accumulated about four times as much radiocalcium and radiostrontium as did the tibia shaft; the $OR_{tissue\text{-}dose}$ values were about the same for both types of bone. The lower $OR_{tissue\text{-}dose}$ values for the ribs in contrast to the tibias are probably real, and reflect the greater ability of the ribs to equilibrate with plasma as the plasma changes with time. After the intravenous dose, the $OR_{tissue\text{-}dose}$ for the incisor tooth of Goat 2 was identical with that of the tibia; thus, the tooth was unable to metabolize these cations differently than certain parts of the skeleton; therefore, this supports the idea that the higher $OR_{tissue\text{-}dose}$ of the tooth after oral administration was related to surface effects.

From the rather constant $OR_{bone\text{-}diet}$ values found in the continuously fed and singly injected goats, it may be implied that, in the long run, the concentration of fallout strontium to total calcium will be identical in all sites of the skeleton. Therefore, "hot spots" in different types of bone or different areas of any bone, as demonstrable with single injections of Ca^{45}, Sr^{90}, or radium *(7, 13)*, will not be formed, at least on a macro-scale. Fallout strontium, being ingested at fairly constant levels over the years, will be uniformly dispersed throughout the bone calcium accreted during that period. Kulp *et al. (8)* have calculated the total skeletal load of Sr^{90} from the analysis of a single type of bone and a correction factor based on the variability with which different types of bone accumulate radiostrontium after a single intravenous injection. This may be applicable to older individuals exposed to fallout Sr^{90} over a relatively short segment of their skeletal history. This correction will be less valid as the length of exposure to Sr^{90} increases, and certainly would not apply to young persons, whose entire skeleton was formed since the time when the biosphere was initially contaminated with Sr^{90}. These considerations have been treated recently by Langham and Anderson *(9)*.

Over-all discrimination. To quantitate the contribution of individual physiological processes to the over-all discrimination, the term strontium-calcium discrimination factor (DF) has been defined as follows *(5)*:

$$OR = (DF_{absorptive})\ (DF_{urinary})\ (DF_{lactational}) \ldots (DF_n) \tag{2}$$

In the lactating animal, it appears that the only processes significantly operative in causing discrimination are absorption, urinary excretion, and secretion into milk. The following equations have been developed, similar to those for discrimination in the pregnant animal *(17)*:

$$DF_{absorptive} = \frac{100 - F_{Sr}}{100 - F_{Ca}} \tag{3}$$

$$DF_{urinary} = \frac{(100 - F_{Ca}) - \left(U_{Sr} \times \dfrac{100 - F_{Ca}}{100 - F_{Sr}}\right)}{B_{Ca} + L_{Ca}} \tag{4}$$

$$DF_{lactational\text{-}body} = \left(\frac{B_{Sr}}{B_{Ca}}\right)\left(\frac{B_{Ca} + L_{Ca}}{B_{Sr} + L_{Sr}}\right) \tag{5}$$

$$DF_{lactational\text{-}milk} = \left(\frac{L_{Sr}}{L_{Ca}}\right)\left(\frac{B_{Ca} + L_{Ca}}{B_{Sr} + L_{Sr}}\right) \tag{6}$$

where

F_{Sr} and F_{Ca} = per cent of ingested dose of Sr^{89} and Ca^{45}, respectively, in feces

B_{Sr} and B_{Ca} = per cent of ingested dose of Sr^{89} and Ca^{45}, respectively, in the body

L_{Sr} and L_{Ca} = per cent of ingested dose of Sr^{89} and Ca^{45}, respectively, in the milk

U_{Sr} and U_{Ca} = per cent of ingested dose of Sr^{89} and Ca^{45}, respectively, excreted in the urine

In derivation, the assumptions are made that the urinary excretion of each element is proportional to the amount absorbed, and that the amount secreted into the milk is proportional to the amount absorbed, minus that excreted into the urine.

Using the mean values (from Table 2), the following are calculated:

$$DF_{absorptive} = 0.24$$
$$DF_{urinary} = 0.60$$
$$DF_{lactational\text{-}milk} = 0.61$$
$$DF_{lactational\text{-}body} = 1.4$$

These DF values then permit a comparison of the importance of the different physiological processes in bringing about the discrimination between calcium and strontium. It is clear that the preferential absorption of calcium from the gastrointestinal tract is of major significance. Even though the total urinary excretion of calcium in the goat is small, this process contributes to the discrimination in an important way; this is because of the much greater plasma clearance of strontium, and because of the relatively small amounts of the two elements that are absorbed and metabolized by the body. The lactation process is of equal importance and tends to reduce the strontium-calcium ratio in the milk; whereas, it raises the strontium-calcium ratio in the body.

ACKNOWLEDGMENTS

The authors wish to express their gratitude for the capable assistance of Dr. I. Kanno, Visiting Scientist from Sendai, Japan, and Mr. Charles Phipps. The assistance of the staff of the animal laboratory, in the care of the experimental animals, is gratefully acknowledged.

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EFFECT OF HEAVY CONCENTRATE FEEDING BEFORE CALVING UPON LACTATION AND MAMMARY GLAND EDEMA [1]

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SUMMARY

Heavy grain-feeding before calving resulted in no increase in the yield of fat-corrected milk during the first 84 days of lactation. Nine cows and nine heifers received, in addition to a basal roughage ration, a supplement of 378 and 273 lb. grain, respectively, during a 6-wk. prepartum period. A similar number of controls received no grain. During lactation all animals were fed roughage and grain according to maintenance and production requirements. Supplemented animals made significantly greater ($P = < 0.01$) increases in live weight but evidenced no significant increase in the incidence or severity of mammary edema as determined by visual ratings and linear measurements. Heifers showed more edema at calving than did older cows.

Thirty years ago, Boutflour *(2)* introduced a method of preparing a cow for lactation which he called "steaming up." In this procedure the cow receives concentrates 6 wk. before the expected date of calving and the allotment is increased weekly. In the final week of the dry period, the amount of grain is increased to 75% of the estimated grain requirement of early lactation.

The stated objects of steaming up are to build up the cow's body reserves and to accustom her to the heavy grain ration she should receive in early lactation. Boutflour *(3)* claimed that milk production in the subsequent lactation is increased by this technique. Although steaming up is a fairly common practice in Britain, it has received little attention from research workers in Britain and no attention from those in the United States. Blaxter *(1)* obtained significantly greater milk yields in early lactation when cows were steamed up for a 6 wk. period by feeding up to 6 lb. of concentrates daily above a control ration consisting entirely of bulky feeds. A third group of cows received supplemental bulky feeds in amounts equivalent in net energy content to the grain fed to the steamed-up group. The milk yields of the third group were increased over the controls, but not significantly.

The effect of a high plane of nutrition compared with a low plane during the dry period also has been studied by Popoff *(9)* and Skrodels *(10)*. These workers obtained increases of up to 70% for the complete lactation as a result of supplementation of the basic dry-period ration with concentrates.

A second aspect of this study arose as a result of the common belief among dairymen that grain feeding during the dry period increases the incidence and severity of mammary edema at calving. However, there is little experimental

Received for publication September 13, 1957.

[1] Data presented in this paper are taken from a thesis submitted by J. F. D. Greenhalgh in partial fulfillment of the requirements for the M.S. degree.

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evidence bearing on the question. Wylie and Hinton *(13)* observed that switching from a plan of normal grain feeding in the dry period to heavy grain feeding in late lactation and no grain in the dry period reduced udder edema and increased production. However, Fountaine, Parrish, and Atkeson *(5)*, in a more thorough investigation, found that the amount of edema at calving was not related to grain feeding during the dry period.

The principal purpose of the present experiment was to examine the effect of steaming up upon the production of cows kept under typical American conditions of feeding and management. Further evidence concerning the effect of such concentrate feeding upon udder condition appeared desirable, in view of the scarcity of studies on the subject and the disagreement in results described above. The problem of measuring accurately the degree of mammary congestion also appeared to be of interest, since no objective method of evaluation of the condition was available.

EXPERIMENTAL PROCEDURE

Animals. Thirty-six cows and heifers of the University dairy herd were employed in the experiment. The five major dairy breeds were represented, but Holsteins predominated. Eighteen cows were placed in two groups of nine with animals of the same breed being equally divided between groups, and 18 heifers were similarly apportioned. Apart from division by breed, the assignment of animals to Group I, Supplemented, or Group II, Control, was at random. All animals were due to calve between October, 1953, and February, 1954.

Feeding and management. Throughout the experiment, all animals were housed in stanchion barns and during lactation were milked by machine at 4:15 A.M. and 3:45 P.M. All feeds were weighed out daily and refusals weighed back. The feeds consisted of good-quality corn silage, medium-quality alfalfa hay, and a 15.4% total protein grain mixture composed of yellow shelled corn, 40, oats, 35, wheat bran, 10, soybean meal, 5, linseed meal, 7.5, steamed bone meal, 1, and trace-mineralized salt 1.5%.

The experimental feeding period began 42 days before each animal was expected to calve. For the whole experiment all animals received daily 2 lb. of silage and 1-½ lb. of hay per 100 lb. live weight. Before calving, grain was fed to the supplemented group only. During lactation all animals were fed according to Morrison's *(8)* optimum standards. Grain was fed in amount sufficient to meet the difference between the TDN provided by the hay and silage, and the calculated requirement. The prepartum and postpartum concentrate feeding schedules are shown (Table 1).

Observations recorded. Milk records were kept for the first 12 wk. of lactation, beginning the fourth morning after calving. Fat tests, according to the Babcock method, were made on milk samples taken from Monday through Thursday each week.

All animals were weighed 42 and seven days before the predicted calving date, and seven and 84 days after calving. Each weight is the mean of three days' weighings carried out at 1 P.M. each day.

TABLE 1
Concentrate feeding schedule

Days prepartum[a]	Daily grain to supplemented group[b]		Days postpartum	Daily grain after calving[c]	
	Cows	Heifers		Cows	Heifers
	——(lb.)——			——(lb.)——	
42–36	5	4	0–4	Discretion of herdsman	
35–29	7	5	5–8	6	4
28–22	9	6	9–12	8	6
21–15	10	7	13–16	10	8
14– 8	11	8	17–88	According to TDN requirement	
7–parturition	12	9			
Av. grain per animal	382	260			

[a] Schedule based on expected calving date.
[b] Control group received no concentrates during dry period.
[c] Grain fed at same rate to supplemented and control animals after calving.

In any study of udder edema, there is the fundamental problem of measuring the amount of swelling and inflammation present. Five methods of measurement were tested by Wilson *(12)*, but in this experiment these were limited to two. In the first method, the severity of the edema was estimated independently by two experienced judges, who examined each animal three days after calving. They rated the udder according to a simple scale, in which 1 represented slight edema and 5 severe edema.

A second method of physical measurement was employed, the principle being to compare the size of the udder at calving with its normal size. Thus, the udder was first measured three days after calving, and further measurements were made 30 days after calving, when a normal condition could be presumed. The measurements, which were made with a tape, were as follows: *a)* udder length: the distance from a mark made on the escutcheon, along the sole of the udder to the junction of the udder with the abdominal wall; *b)* udder width: the distance from the junction of the udder with the abdomen on one side to the same point on the other side, the tape being placed midway between the front and rear teats.

These measurements were made at 8:30 A.M., 4 hr. after milking. Some difficulty was experienced in finding a suitable agent for making a mark on the escutcheon, and the substance eventually employed was red fingernail-varnish.

Statistical analysis. In the original design of the experiment it was planned that there should be two treatment groups, each containing two subclasses; that is, a supplemented group divided into subclasses of nine cows or nine heifers, and a control group similarly divided. However, the failure of one cow to calve, and the loss of live weight data resulting from early calving, led to the formation of disproportionate subclass numbers. Consequently, the necessary analyses of variance were made by the method of fitting constants *(6)*.

RESULTS

The average length of the prepartum feeding period was 43 days for cows and 40 days for heifers. Feed refusals throughout the trial were negligible. Milk yields, live weight changes, and mammary edema measurements recorded during the experiment are shown (Table 2). In this table, direct comparisons between

TABLE 2

Milk yields, live weight changes, and mammary edema measurements

	Measurement	Means				Residual standard deviation	Differences	
		Supplemented		Control				
		Cows	Heifers	Cows	Heifers		Supplemented vs. control	Cows vs. heifers
I	Milk yields							
	Daily yield of FCM in first 84 days of lactation *(lb.)*	47.5 (9)[a]	39.4 (9)	49.4 (8)	38.3 (9)	± 8.1	− 0.1	9.6[b]
	Daily yield of actual milk in first 84 days of lactation *(lb.)*	46.8 (9)	41.3 (9)	49.5 (8)	40.4 (9)	± 9.7	− 0.7	7.3[c]
II	Live weight changes							
	Between 42 and seven days prepartum *(lb.)*	+67.8 (9)	+80.4 (7)	+43.0 (4)	+42.7 (7)	±29	30.5[b]	− 1.4
	Between seven and 84 days postpartum *(lb.)*	−43.9 (9)	−17.2 (9)	−24.3 (8)	+ 2.0 (9)	±41	20.1	27.1
III	Mammary edema measurements							
	Visual rating (1 = slight, 5 = severe edema)	1.86 (8)	3.61 (9)	2.35 (7)	3.09 (9)	± 0.84	− 0.02	− 1.26[b]
	Udder length measurement. Reduction in length from three to 30 days postpartum *(in.)*	2.58 (6)	4.14 (7)	1.79 (7)	3.44 (9)	± 2.02	0.70	− 1.60[c]
	Udder width measurement. Reduction in width from three to 30 days postpartum *(in.)*	2.42 (6)	3.21 (7)	0.93 (7)	2.33 (9)	± 2.55	1.13	− 1.10

[a] Figures in parentheses are numbers of animals.
[b] Significant at the 1% level of probability.
[c] Significant at the 5% level of probability.

subclass means are permissible, since all interactions were nonsignificant. The subclass standard deviations were tested for homogeneity and were found to be combinable.

As one would expect, the FCM production of mature cows was significantly greater ($P = <0.01$) than that of first-calf heifers. The differences between the supplemented and the control groups in their production of both actual milk and FCM during the first 84 days of lactation were nonsignificant. Production curves drawn by plotting yield against time did not suggest that differences between groups would develop later in the lactation.

As shown (Part II of Table 2), the mean live weight gain for nine supplemented cows from 42 to seven days prepartum was 67.8 lb., and for four control cows, 43.0 lb. Gains for seven supplemented heifers averaged 80.4 lb., compared with 42.7 lb. for seven controls. Group I animals made significantly greater weight gains ($P = <0.01$) during the dry period. After calving the intra-class variability was great and mean differences were nonsignificant.

Results of the edema study appear in Part III of Table 2. Assessment of the severity of the condition at calving by means of judges' ratings showed a highly significant difference between cows and heifers, but no difference as a response to treatment. Measurements of the decrease in udder length between three and 30 days postpartum again indicated differences between age groups, but at a lower level of significance. The measurements of the reduction in udder width showed no significant differences between ages or treatments.

The agreement between methods of measuring edema was tested by calculating a number of simple and multiple correlation coefficients, which are shown (Table 3).

TABLE 3

Comparison of the different methods of measuring udder edema

Correlation between:	"r" value
1. The ratings of one judge and those of the other, $r_{x_1 \cdot x_{11}}$	0.8201[a]
2. Ratings and udder length decreases, $r_{x \cdot y}$	0.5123[a]
3. Ratings and udder width decreases, $r_{x \cdot z}$	0.1654
4. Ratings and both measurements, $r_{x \cdot yz}$	0.5138[b]
5. Udder length decreases and udder width decreases, $r_{y \cdot z}$	0.3919[b]

[a] Significant at 1% level of probability.
[b] Significant at 5% level of probability.

This table shows that the judges agreed well when making their independent estimates of the amount of edema present. When the measurements were compared with the ratings, the decrease in udder length between three and 30 days postpartum appeared to evaluate the condition much better than the decrease in udder width in the same period.

DISCUSSION

Milk yields and live weight changes. The feeding of a supplementary grain ration prior to calving could, conceivably, bring about a subsequent increase in milk production in a number of ways. The first and most obvious mode of

action is that deposits of body fat laid down in the dry period could be "milked off" during lactation. Secondly, there is the possibility that an improved supply of nutrients in late pregnancy could encourage extra development of milk secretory tissue; hence, an augmented milk flow. Finally, steaming up could improve milk production by what might be called a metabolic preparatory effect. It is generally considered advisable to increase postpartum feed intake gradually, to avoid trouble from digestive disorders and, possibly, milk fever, with the result that nutrient intake may fall behind nutrient requirements. Consequently, at a time when milk production should be stimulated, a brake is placed on secretion. However, as a result of accustoming the cow to heavy grain feeding before calving, the build-up of nutrient intake could be accelerated, and the cow could be brought up to full feed soon after calving.

Results of this experiment provide no evidence that any of these modes of action was operating. Although supplemented animals made significantly greater weight gains before calving and appeared to lose this weight advantage after calving, their milk yields were not increased over the controls. It is possible that the method of feeding after calving suppressed any effects of steaming up. All animals may have been fed so liberally that the supplemented animals had no opportunity to make efficient use of their extra weight increases. This is a matter for conjecture, but it is suggested that the conversion of feed to milk via the intermediate step of body fat is likely to be an unprofitable procedure. The third suggested mode of action, the metabolic preparatory effect, may not have had a chance to manifest itself. The experimental animals were not brought on to full feed immediately after calving, but the build-up was faster than is normally the case (see Table 1).

Before turning to other experiments in which a high plane of nutrition during the dry period has proved its worth, it must be stressed that steaming up, as promoted by Boutflour *(3)*, involves the supplementing with grain of normally adequate rations. Consequently, when measuring the effect of steaming up, it is important that control animals should be fed at such a level that they can make live weight increases commensurate with the growth of the fetus and accompanying tissues, i.e., controls must not actually lose flesh during the dry period. In New Zealand *(4, 7)*, high and low planes of nutrition during the dry period were compared by their effect on the following lactation. In these experiments, the control animals were made to lose weight prior to calving, and so the milk-yield differences represent a response to adequate nutrition compared with underfeeding. Steaming up, on the other hand, is overnutrition and its true value can be assessed only by comparing it with adequate nutrition.

In the investigation of steaming up conducted by Blaxter *(1)*, cows whose rations were supplemented either with grain or with bulky foods gained approximately 80 lb. per head during the period 42 to seven days prepartum. Controls gained only 13 lb. per head, an amount which represents an actual loss of flesh when the increase in weight of the fetus and its membranes is taken into account *(11)*. Consequently, steaming up was being compared with underfeeding. In the experiment reported here, however, supplemented animals gained an

average of 73.3 lb. and controls 42.8 lb., from 42 to seven days prepartum. The latter weight increase indicates that the controls were being fed an adequate ration.

The present experiment demonstrates the effect of steaming up on milk production, in an area in which cows are usually well-fed during the dry period.

Mammary edema. The first aim in measuring the udder edema encountered in this experiment was to compare objective and subjective methods, with a view to establishing a satisfactory objective method of evaluating the condition. The second aim was to investigate any connection between prepartum grain feeding and mammary edema.

It is logical to assume that an over-all estimate of the degree of swelling in an irregularly shaped body should be more accurate than any one linear measurement; this assumption is confirmed by the results of this experiment. Judgment by eye appeared to be a more satisfactory method of assessing the severity of the condition than measurements of udder length and width. Thus, Table 3 shows that the correlation between the ratings of one judge and those of the other was far closer than any of the other correlations investigated. This conclusion does not solve the problem of finding an objective measurement, but it does suggest that for any one experiment judgment by the practiced eye is a fairly satisfactory method of measuring edema.

The length measurement of the udder appeared to be more useful than the width measurement in evaluating the edema. The apparent reason for this is that length measurements included the accumulation of fluid often seen at the fore end of the udder, whereas width measurements did not. When measurement changes from three to 30 days postpartum were expressed on a percentage basis, and differences in basic udder size between cows were eliminated, the correlation between the measurements and judges' ratings was not increased.

The symptoms of mammary edema are well known, but their cause is still a matter for speculation. Although the relation between prepartum grain feeding and an accumulation of lymph in the udder is not obvious, dairymen often believe that such a connection exists. However, in this experiment, feeding as much as 9 or 12 lb. of grain per day before calving did not increase the amount of edema. Singificant differences occurred between cows and heifers, but there were no interaction effects, i.e., there was no evidence that grain feeding increased edema in cows but not in heifers, or vice versa. The difference in severity of edema due to age of animal, in this case, supports the observations of practical dairymen.

ACKNOWLEDGMENT

The authors gratefully acknowledge the assistance of the following: W. W. Yapp and E. E. Ormiston for judging the severity of udder edema; R. W. Touchberry for advice on the statistical treatment of the results, and R. L. Shirley and his staff for their care of the experimental animals.

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RELATIVE MERITS OF FIVE MEASURES OF A DAIRY SIRE'S TRANSMITTING ABILITY [1, 2]

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SUMMARY

Lactation milk and fat records of 6,949 daughters, 6,201 different dams, and the contemporary D.H.I.A. herd averages of 2,420 herds were studied, to compare five measures of a dairy sire's transmitting ability. They were the daughter average, daughter-dam difference, equal-parent index, daughter-contemporary herd difference, and daughter-contemporary-herd index. The daughters and dams were included in the D.H.I.A. proofs for 350 Guernsey and 486 Holstein sires used in artificial-breeding associations, and whose proofs were published for 1949–1951. A variance component analysis did not reveal any marked superiority for any of the measures, as determined by the percentage of the total variance comprised by the sire component for each of the measures. Sufficient data for the Holsteins were available for an empirical check of the reliability of these five measures. The simple daughter average appeared to be about as reliable as the equal-parent index or the daughter-herd index to predict future daughters' production for milk or fat. The D.H.I.A. herd average included the daughter in question and reduced the magnitude of the sire component for the herd measures. Use of a contemporary-herd average, which excludes the daughter in question in the two daughter-herd measures, should improve their merit as measures of a dairy sire's breeding value compared to the other measures in this study.

The performance of later daughters of sires selected for use in artificial-breeding associations, on the basis of proofs in one or two herds (natural), usually has reflected little of their sire's original apparent superiority *(10, 11)*. Much of the disappointment in the performance of the artificially sired daughters undoubtedly has resulted because conventional methods of evaluating sire proofs have not adequately accounted for the influence of herd differences and changes in intra-herd environment. The use of the contemporary herd average has been suggested to avoid this glaring weakness *(2, 8, 21)*. Most of the differences between herds are environmental *(15, 18, 19)* and, presumably, most of the intra-herd environmental changes are reflected by the herd average. Inasmuch as the herd average is readily available, could it be substituted for the dam's average in the daughter-dam difference and in the equal-parent index? If so, many more daughter-herd comparisons than daughter-dam comparisons would be permitted, since about one-half of the tested daughters have tested dams.

For these reasons, five measures of a sire's transmitting ability for milk and fat production, namely, the daughter average ($\bar{O}$), daughter-dam difference

Received for publication December 27, 1957.

[1] Contribution from the North Carolina Agricultural Experiment Station and published with the approval of the Director of Research as Paper No. 872 of the Journal Series.

[2] The data in this paper were taken from a dissertation presented by the senior author to the Graduate School, North Carolina State College, in partial fulfillment of the requirements for the degree of Doctor of Philosophy, 1955.

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($\overline{O}$-$\overline{D}$), equal-parent index ($2\,\overline{O}$-$\overline{D}$), daughter-contemporary herd difference ($\overline{O}$-$\overline{H}$), and daughter-contemporary herd index ($2\,\overline{O}$-$\overline{H}$) were studied. The latter two measures can be visualized simply by substituting the actual herd average for the dam's average production in the daughter-dam difference and the equal-parent index.

EXPERIMENTAL PROCEDURE

Data. The data included lactation milk and fat records of 6,949 daughters, 6,201 different dams, and contemporary D.H.I.A. herd averages for 2,420 herds. The daughters and dams were included in the D.H.I.A. proofs of 350 Guernsey and 486 Holstein sires used in artificial-breeding associations in the United States, whose proofs were published for the years 1949–1951.

To be included in this study, each daughter had to have a contemporary D.H.I.A. herd average for at least one lactation and a dam with a record. The contemporary herd average used was the herd average completed the year following the year the individual cow freshened. This average was chosen after considering that many herds are on a reporting year which does not coincide with the calendar year, that the cow makes a record 10 mo. after freshening, and that the D.H.I.A. average is listed according to the year in which it is completed. Except for the D.H.I.A. herd average, all records were adjusted to a 305-day, 2 × milking, mature equivalent (M.E.) basis.

Naturally and artificially sired daughters of the sires were analyzed separately. The average production of the four groups studied is shown (Table 1).

TABLE 1
Production averages of groups in study

Group[a]	Daughters	Sires[b]	Herds	305 day, 2 ×, M. E.		
				Milk	%	Fat
G–N	1,865	636	558	8,677	4.93	426
G–A	438	324	276	8,066	4.89	392
H–N	3,582	1,239	1,078	12,901	3.68	472
H–A	1,064	795	672	12,210	3.59	436

[a] G–N, Guernsey natural; G–A, Guernsey artificial; H–N, Holstein, natural; H–A, Holstein, artificial.

[b] These represent the number of sires on a within-herd and state basis. A total of 836 different sires was included, 350 Guernseys and 486 Holsteins.

The lower production level for the artificially sired daughters, as compared to the naturally sired animals, is typical of the situation throughout the country as reported from D.H.I.A. summaries *(11)*.

Analysis. Each of the five measures was expressed as a linear equation or model which included the effects of herd, sire-within-herd, and within-sire and herd effects on an intra-state basis. The sire-by-herd effect was not included, since previous analysis of similar data had shown it to be small *(13)* and the sire-by herd interaction variance could not be estimated with reasonable precision from these data. Using these models, the variances for each of the measures were expressed in terms of the components of variance for the simple daughter average. The details of procedure are described elsewhere *(6)*, but the resulting

expressions for the variances, pertinent to the interpretation of later results, are as follows:

(1) $$\sigma^2_{(\bar{o})} = \sigma^2_s + \frac{\sigma^2_h}{\text{No. of herds}} + \frac{\sigma^2_e}{\text{No. of daughters}}$$

(2) $$\sigma^2_{(\bar{o}-\bar{p})} = \sigma^2_s + \frac{2\,\sigma^2_h\,(1-r)}{\text{No. of herds}} + \frac{\sigma^2_e + \sigma^2\theta}{\text{No. of daughters}}$$

(3) $$\sigma^2_{(2\,\bar{o}-\bar{p})} = 4\,\sigma^2_s + \frac{\sigma^2_h\,(5-4r)}{\text{No. of herds}} + \frac{2\,\sigma^2_g + 5\,\sigma^2\theta}{\text{No. of pairs}}$$

(4) $$\sigma^2_{(\bar{o}-\bar{n})} = \sigma^2_s\left(1 - \frac{2n_{ii.}}{n_{i..}} + \frac{n^2_{ii.}}{n^2_{i..}}\right) +$$

$$\frac{\sum_{ij \neq i} 1\,\frac{n_{ij.}{}^2\,\sigma^2_s}{n^2_{i.}} + 2\,\sigma^2_h\,(1-r)}{\text{No. of herds}} + \frac{\sigma^2_e\left(1 - \frac{1}{n_{i..}}\right)}{\text{No. of daughters}}$$

(5) $$\sigma^2_{(2\,\bar{o}-\bar{n})} = \sigma^2_s\left(4 - \frac{4n_{ii.}}{n_{i..}} + \frac{n^2_{ii.}}{n^2_{i..}}\right)$$

$$\frac{\sum_{ij \neq i} 1\,\frac{n^2_{ij.}\,\sigma^2_s}{n^2_{i..}} + \sigma^2_h\,(5-4\,r) + \frac{\sigma^2_e\left(4 - \frac{3}{n_{i..}}\right)}{\text{No. of daughters}}}{\text{No. of herds}}$$

Where: σ_s^2 denotes the variance due to the additive genetic effects of the sire, possibly some epistatic effects (σ_i^2) and genetic environmental interactions.

σ_h^2 denotes the variance due to the genetic and environmental effects common to a herd but different from these effects on other herds.

σ_e^2 denotes the variance peculiar to the cow. This includes existing errors in measurement of the record and environmental influence not accounted for by the herd effect (σ^2) and the genetic effects not accounted for by the sire. Hence, $\sigma_e^2 = \sigma_\iota^2 + \sigma_l^2 + \frac{3\,\sigma_g^2}{4} + \sigma^2$ where σ_d^2 represents the dominance variance.

σ_g^2 denotes the variance due to differences in the additive breeding value of the individual cow and, as used, possibly includes some epistatic variance and genotype environmental interaction.

σ_θ^2 differs from σ_e^2 in that it does not include the additive genetic variance attributed under random mating to the dam and to chance at segregation ($\sigma_\theta^2 = \sigma_d^2 + \sigma_i^2 + \sigma^2$).

r is the correlation between the herd effects for the daughter and her dam or herd mates.

$n_{ii.}$ is the number of daughters of the sire under consideration in the i^{th} herd.

$\sum_{ij \neq 1} n_{ij.}$ is the number of daughters of other sires in the i^{th} herd.

Individual analyses of variance were computed for each of the five measures for milk and fat production for the two breeds, divided into natural and artificial groups. From each, estimates of the variance components were derived for herd, sire-within-herd, and within-herd and sire effects on an intra-state basis using Method I of Henderson *(7)*.

Since averages of records, differences, and indexes were used in the several measures, it was necessary to adjust the error variances to a single record basis, so that the results could be presented independent of the numbers of records per animal characteristic of these data. The formula used to adjust the error variance of an average of n records was

$$\sigma_e^2 = \sigma_e^2 \frac{\overline{n}}{1 + (\overline{n} - 1)\, r_{dd}}$$

where $\overline{n}$ is the harmonic mean of the number of records per cow. The repeatability (r_{dd}) of successive records of a cow was taken as 0.50 for milk and 0.40 for fat production, as based on values from the literature *(1, 9, 12, 14, 20)*. The harmonic means in these data were as follows:

	Daughters	Dams
Guernsey, natural	1.32	2.18
Guernsey, artificial	1.22	2.08
Holstein, natural	1.32	2.27
Holstein, artificial	1.22	2.14

Similarly, the error variances for the other measures were adjusted to a single record or comparison basis with appropriate formulae being derived for each.

The fraction of the total estimated variance in each measure, which was represented by the sire component estimate, was computed. Since some of the sires had both naturally and artificially sired daughters, an empirical check was obtained of the values from the variance component analysis. Correlations for each of the five measures of a sire's transmitting ability were computed between naturally and artificially sired daughters and between a specific number of first and later artificially sired daughters for the Holstein data.

RESULTS AND DISCUSSION

Variance component analysis. Since the individual lactation milk and fat records are basic to the interpretation of the other measures, the importance of the several sources of variation in them is of primary concern. The components of variance for milk and fat for the daughter average are given in the second,

third, and fourth columns of Table 2. The percentages of the intra-state variance which they represent are given in the second column of Table 3.

Results comparable to those obtained in this study were secured by Legates *et al. (13)*, in an analysis of records of 5,234 Guernsey and 17,581 Holstein daughters of sires used in artificial-breeding associations. The Guernsey sire components accounted for 5.8 and 4.1% of the intra-state variance in milk and fat, respectively, in their data. The comparable percentages for the Holstein sire components were 7.7 and 7.8, respectively. In a study of New York Holstein data *(5)*, the sire component accounted for only 2.8% of the variance in lactation fat records. Carter *(3)* has since reported that adjustments for year, herd, and season effects removed much nongenetic variation. Following these adjustments, the sire component accounted for 7.7% of the variation.

The differences in the results for the Guernseys and the Holsteins in this study possibly could be explained by sampling, since the number of Guernsey sires was considerably less than the number of Holstein sires. However, lower valúes for the Guernsey sire components were also obtained in the study by Legates *et al. (13)*.

Values for the herd component of variance are in general agreement with those in the literature *(13, 16, 17)*. Herd differences generally accounted for a larger percentage of the total variance for the artificial groups.

Estimates of the variance components for all five measures for milk and fat are given (Table 2). The variances which are estimated are the functions of the components presented in the investigation for each measure, *e.g.*, for the daughter-dam difference, the σ_h^2 as computed estimates the expected $2\,\sigma_h^2\,(1\text{-}r)$. Inspection of the values (Table 2) reveals several circumstances which could be predicted on the basis of the formulae derived for the variances of the measures. The sire variance for the equal-parent index is about four times as large as the corresponding components for the daughter average; likewise, the sire variance for the daughter-herd index is about four times the corresponding component for the daughter-herd difference. The sire variances for the two measures using the herd average were consistently smaller than they were for their counterparts, the daughter-dam difference and the equal-parent index. This was to be expected, since the cow's own record was a part of the herd averages used.

The sire component for the daughter-dam difference was expected to be comparable in size to the component for the daughter average. Values for the daughter-dam difference are higher for the Holsteins. Time trends during the interval the daughters and dams made their records could have contributed to this; but, if they were present, they should have influenced the equal-parent index to as great an extent. The small number of degrees of freedom for sires, however, does not rule out the possibility that sampling may be responsible.

One criterion for evaluating a measure of a sire's transmitting ability is to require the measure to maximize sire differences, thereby permitting more accurate choices among sires. The extent to which this is accomplished can be expressed in terms of the fraction of the total variance which is represented by

TABLE 2

Estimates of variance components for all five measures

Group	Daughter			Daughter-dam			Equal-parent index			Daughter-herd			Daughter-herd index		
	Herd	Sire	Error[a]	Herd	Sire	Error[a]	Herd	Sire	Error[a]	Herd	Sire	Error[a]	Herd	Sire	Error[a]
							Milk								
G–N	9,734	890	20,535	5,722	2,418	37,096	24,662	4,502	94,732	4,414	525	20,628	19.338	2,633	81,641
G–A	3,067	9,701	15,136	3,317	7,685	33,099	−2,302	37,718	81,154	−20	10,969	15,357	−2,055	40,157	60,481
H–N	18,871	8.321	42,561	5,870	13.673	78,314	39,429	32,726	199,197	1,078	8,740	44,060	21,615	30,940	171.823
H–A	21,304	7,045	31,509	11,699	13,568	53,608	50,809	38,647	138,725	8,200	6,022	32,761	40,098	25,459	127,490
							Fat								
G–N	2,588	344	4,830	1,400	561	8,996	6.441	1,285	22,822	841	262	4,861	4,592	1,161	19,221
G–A	2,371	594	3,647	1,950	28	7,823	4,257	3,180	19,293	1,691	992	3,576	5,809	2,905	13,322
H–N	2,717	990	5,946	961	1,493	11,066	6,060	3,552	28,078	1,392	771	6,071	3,590	3,045	23,834
H–A	3,017	806	4,191	1,451	1,173	7,824	6,837	3,652	19,839	996	710	4,254	5,527	2,908	16,748

[a] Within-sire, within-herd—among paternal sisters in the same herd.

the sire component. Table 4 presents such a comparison for all five measures. Close inspection of this table reveals that no measure has emerged as superior. Although the percentages for the Guernsey natural group are markedly lower than those for the other three groups, there is no consistent trend in any one measure either for milk or for fat.

Using the variance components, repeatability values for each measure can be computed for any number of daughters or herds. For example, the repeatability of the daughter average

$$\sigma_s^2/\sigma_s^2 + \frac{\sigma_h^2}{\text{No. of herds}} + \frac{\sigma_e^2}{\text{No. of daughters}}$$

for fat for a Holstein sire with five artificially sired daughters in five herds is 0.36 or, for ten daughters in ten herds, 0.53. Usually, natural proofs include daughters in just one or two herds. For a Holstein sire with five daughters all in one herd, the repeatability of the daughter average for fat is 0.20 or for a sire with ten daughters all in one herd it is 0.23. In each case these values indicate the fraction of the superiority of a bull's first daughters that would be expected to show in a later group of daughters located in different herds. For the other measures considered, such values represent the fraction of the superiority based on that particular measure that one would expect to be exhibited by a later group of progeny, evaluated with that measure.

Much hope has been expressed for the two herd measures. Perhaps their sire components might have represented higher percentages of the total variance if the most contemporary herd average could have been secured in all cases.

Since the cow's own record could not be separated from the herd average, herd size also introduced another important factor. For herds smaller than the average of 24.2 cows for Holsteins and 26.4 cows for Guernseys in this study, the sire variance of these measures is reduced at an increasing rate as the herd size diminishes; whereas, the sire variance is reduced little for herds larger than those in the study. This is shown by the repeatability estimates for ten daughters in ten herds for the daughter-herd difference for fat production of the Holstein artificial group for 10, 20, 30, and 40 cow herds of 0.44, 0.54, 0.56 and 0.57, re-

TABLE 3

Percentage of the total variance which is represented by the sire component for each of the measures

Breed group	Daughter	Daughter-dam	2 Daughter-dam	Daughter-herd	2 Daughter-herd
			Milk		
G–N	2.9	5.3	3.6	2.1	2.5
G–A	34.8	17.4	31.7	41.7	39.9
H–N	11.9	14.0	12.1	16.2	13.8
H–A	11.8	17.2	16.9	12.8	13.2
			Fat		
G–N	4.4	5.1	4.2	4.4	4.6
G–A	9.0	.3	11.9	15.8	12.6
H–N	10.2	11.0	9.4	9.4	10.0
H–A	10.1	11.2	12.0	11.9	11.5

spectively. With the use of a contemporary herd average which excludes the daughter in question, the two daughter-contemporary-herd measures would be likely to give higher percentage values for the sire component than those in this study.

Empirical analysis. Correlations between each of the five measures of a sire's transmitting ability based on naturally or artificially sired daughters, and the performance of a specific number of future artificially sired daughters, were computed. For this analysis, all five of the naturally sired daughters were from the same herd, and each of the artificially sired daughters or comparisons for a sire were from a different herd. With these restrictions, only the Holstein groups had sufficient data to warrant analysis. Correlation values obtained from this phase of the study are given (Table 4).

While the correlations do not show a decisive margin in favor of any one of the measures, the simple daughter average appears to be almost as reliable as any of the other measures. The correlation values involving the daughter average, the equal-parent index, and the daughter-herd index are of approximately the same magnitude. The correlations for the daughter-herd difference were generally slightly lower than these three; whereas, those for the daughter-dam difference were lowest in seven of the eight comparisons. Use of a nonpaternal stablemate's average instead of the herd average probably would improve the merits of the two daughter-herd measures, because in the average- and small-sized herds a significant portion of the sire effect is removed when the herd average is subtracted from the daughter average.

The empirical correlations between the original natural proof and a later artificial proof were consistently lower than those for an original and a later artificial proof for the one comparable situation. This has been the experience of many in artificial breeding *(10)*. There was no real difference in the ranking of the measures between the artificial and the natural groups.

These results are in partial agreement with those of Carter *et al. (4)*. They found the daughter-dam difference and a daughter-contemporary difference to be

TABLE 4

Correlations between the measures of a sire's transmitting ability and the performance of his artificially sired daughters (Holsteins)

Variables[a]	No. of sires	Measure				
		Daughter	Daughter-dam	2 Daughter-dam	Daughter-herd	2 Daughter-herd
				Milk		
X_1 and X_2	39	0.20	−0.11	0.18	0.18	0.19
X_2 and X_3	47	0.28	0.08	0.30*	0.21	0.28
				Fat		
X_1 and X_2	39	0.26	0.15	0.24	0.27	0.32
X_2 and X_3	47	0.34*	0.16	0.35*	0.40**	0.38**

* Denotes statistical significance at 5% level of probability.
** Denotes statistical significance at 1% level of probability.
[a] X_1 = First five naturally sired daughters.
X_2 = First five artificially sired daughters.
X_3 = Next five artificially sired daughters.

about equal in merit. However, in their study the daughter average appeared to be of little value, and the equal-parent index was slightly less dependable than the daughter-dam and the daughter-contemporary comparisons.

ACKNOWLEDGMENTS

The authors wish to express their appreciation to F. J. Verlinden and the staff of the Statistical Laboratory at North Carolina State College. They also wish to acknowledge the assistance of J. F. Kendrick of the Dairy Herd Improvement Section of USDA in assembling the data for the study.

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EFFECTS OF THYROTROPHIN, L-THYROXINE, AND L-TRIIODOTHYRONINE ON LEVELS OF PLASMA PROTEIN-BOUND IODINE IN DAIRY BULL CALVES[1, 2]

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SUMMARY

Intramuscular injections of thyrotrophin and intravenous injections of sodium L-thyroxine pentahydrate and sodium L-triiodothyronine were made in dairy bull calves (body wt. from 166 to 264 kg.), and plasma levels of protein-bound iodine (PBI) were used as a response criterion. The injection of thyrotrophin (Armour) in amounts of 10, 20, 40, and 80 U.S.P. units caused maximum PBI increases above initial levels ranging from 3.05 to 8.02 μg. % at 15 to 31 hr. postinjection. The mean biological half-life of the endogenously produced PBI, during its return to normal levels, was 22.1 hr. L-thyroxine (2.5 and 5.0 mg.) and L-triiodothyronine (5.58 and 11.16 mg.) were injected in each of three trials. Two first-order exponential regressions characterized the decline of PBI to normal in each instance. The first represented the rate of movement of the materials into their respective volumes of distribution (mean biological half-time of 1.93 hr. for L-thyroxine and 0.31 hr. for L-triiodothyronine). The second regression measured the rate of peripheral utilization or disposal of the materials (mean biological half-life of 18.0 hr. for L-thyroxine and 5.10 hr. for L-triiodothyronine). The mean volume of distribution for L-thyroxine was 9.07% of body wt.; whereas, the corresponding value for L-triiodothyronine was 37.5%. The constants developed in the L-thyroxine infusion trials were used to estimate the rate of thyroxine secretion or degradation (mean of 0.231 mg. per day per 100 lb. body wt.).

The thyroid gland accumulates iodide from the blood *(13, 16)*, synthesizes thyroglobulin *(2)*, and either stores thyroglobulin in the follicles of the gland or discharges thyroxine *(16)* into the circulation, all as the result of the activity of thyrotrophin from the anterior pituitary.

It has been determined that thyroxine is the major form of protein-bound organic iodine in the normal plasma *(2, 12, 15)* and in plasma following administration of thyrotrophin *(6)*. Both endogenous and exogenous thyroxine in the plasma are protein-bound in the same way, and have identical protein linkage *(5)*. Exogenous triiodothyronine also becomes protein-bound in the plasma *(3)*.

Rate constants have been presented for the disappearance of thyrotrophin-stimulated protein-bound iodine (PBI) *(10)* and, also, for the disappearance of plasma PBI resulting from exogenous thyroxine *(9, 14)*. In addition, Ingbar and Freinkel *(7)* have presented a method for estimating the daily rate of thyroxine degradation which is equivalent to thyroxine secretion rate. This may be calculated utilizing as components the normal level of plasma protein-

Received for publication January 30, 1958.

[1] Paper of the Journal Series, New Jersey Agricultural Experiment Station, Rutgers University, the State University of New Jersey, Department of Dairy Science, New Brunswick.

[2] This study was part of a Northeast Regional Project (NE-30, The Relation of Certain Physiological Factors to the Productive Capacity of Dairy Cattle), a cooperative study involving agricultural experiment stations in the Northeastern Region and supported in part by regional funds of the United States Department of Agriculture.

bound iodine, the fractional rate of turnover per day of extrathyroidal thyroxine, and the thyroxine distribution volume or space.

It is the purpose of this paper (using dairy bull calves) to present the quantitative aspects of the stimulation of the thyroid as reflected in plasma level of PBI; to characterize the rate of disappearance or turnover rates of PBI from plasma after the intravenous injections of L-triiodothyronine and L-thyroxine, and to estimate the daily thyroxine secretion or degradation rates, using the principles proposed by Ingbar and Freinkel *(7)*.

MATERIALS AND METHODS

Two dairy bull calves (Guernsey 200[6] and Holstein 639[6]) were used in a series of separate experiments in which thyrotrophin (TH) was injected intramuscularly and L-thyroxine and L-triiodothyronine were injected intravenously. The thyrotrophin weighed 1.515 mg. per U.S.P. unit.[3] The L-thyroxine was in the form of sodium L-thyroxine pentahydrate[4] (mol. wt.—888.995, purity—96.2%); whereas, the L-triiodothyronine was in the form of sodium L-triiodothyronine[5] (mol. wt.—673.003, purity—92.4%).

Blood samples were taken initially and at varying intervals of time following the injection of the above materials and were assayed for plasma PBI by the method of Brown *et al. (1)*, as modified by Lennon and Mixner *(8)*. The PBI responses were calculated by subtracting the initial level of PBI from the subsequent levels of PBI in any given experiment.

RESULTS AND DISCUSSION

Thyrotrophin. In successive experiments, G200[6] was injected intramuscularly with 10 and 40 U.S.P. units of thyrotrophin and H639[6] was injected with 20 and 80 U.S.P. units of thyrotrophin. The nature of the PBI responses, as a function of the time after injection of the thyrotrophin, is presented as a semilogarithmic plot (Figure 1). In three of the four trials, maximum levels of PBI were reached at about 15 hr. after injection, and thereafter declined at a rather steady rate.

The quantitative aspects of the thyrotrophin-PBI response curves (Figure 1) may be analyzed by the methods outlined by O'Neal and Heinbecker *(10)* for thyrotrophin stimulation of PBI in hypophysectomized dogs. These analyses are summarized (Table 1). It is suggested that the declining phase of PBI from the plasma is owing entirely to the disappearance of PBI from the plasma and is best described by a simple exponential regression having the general formula, $PBI = ae^{-bt}$. This regression may be calculated by the least squares method, in which X-values are time in hours after injection of thyrotrophin and Y-values are the natural logarithms of the PBI values in μg. %.

[3] Thyrotrophin, Lot R491031, was generously supplied by Dr. Irby Bunding, The Armour Laboratories, Chicago.

[4, 5] Supplied through the courtesy of Dr. H. L. Saunders, Smith, Kline and French Laboratories, Philadelphia.

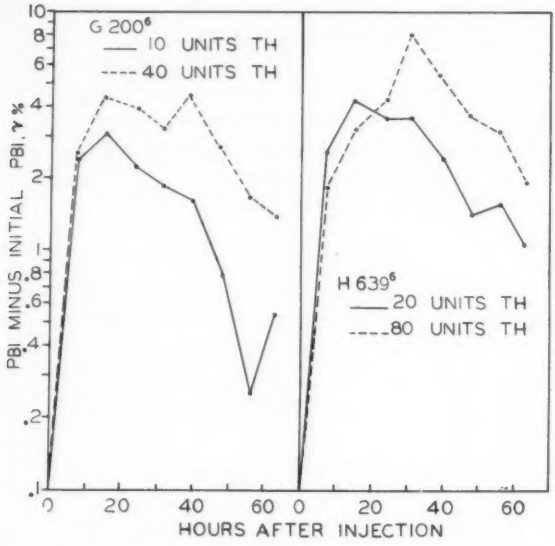

FIG. 1. Semilogarithmic plot of the PBI responses of dairy bull calves injected intramuscularly with varying doses of thyrotrophin (Armour).

TABLE 1

Summary of certain PBI response characteristics to thyrotrophin injected intramuscularly in two dairy bull calves

Characteristic	Animal				
	G200[6]		H639[6]		Means
Age *(mo.)*	5.4	6.1	4.9	5.6	
Body weight *(kg.)*	166	188	189	218	
Normal PBI *(μg. %)*	3.06	4.23	3.36	4.08	3.68
Dosage TH *(U.S.P. units)*	10	40	20	80	
Maximum stimulation of PBI above normal level *(μg. %)*	3.05	4.38	4.22	8.02	
Time to reach maximum PBI *(hr.)*	15±4	15±4	15±4	31±4	19
PBI disposal curve					
1. Degrees of freedom *(n−2)*	8	5	8	3	6
2. Coefficients of correlation between time in hours after thyrotrophin administration and the natural logarithms of PBI *(μg. %)*	−0.93[a]	−0.88[a]	−0.94[a]	−0.99[a]	−0.94[a]
3. Intercept of curve on Y-axis at zero-time *(PBI-μg. %)* (a)	7.86	7.48	6.11	28.39	
4. Calculated total amount of thyroxine stimulated by TH, based on thyroxine space or volume of distribution of 9.07% of body wt. *(mg.)*	1.812	1.953	1.604	8.596	
5. Slope of rate constant of regression of PBI to normal levels $(-b_2)$ *(per hour)*	0.0485	0.0244	0.0240	0.0416	0.0346
6. Half-life of PBI [*hr.* $(t_{1/2})$]	14.3	28.4	28.9	16.7	22.1

[a] Significant at the 1% level ($P < 0.01$).

By use of the regression and its associated constants, five characteristics concerned with PBI disposal from the plasma may be calculated (Characteristics 2 to 6, Table 1).

Characteristic 2 is the coefficient of correlation *(r)* between the time in hours after thyrotrophin injection (*X*-variable) and the natural logarithms of the PBI levels in μg. % (*Y*-variable) during the declining or disposal phase of PBI response. The mean coefficient of correlation for the four trials was −0.94 (Table 1), which is considered to be satisfactorily high.

The *a* constant of the general equation (Characteristic 3, Table 1) is the *Y*-axis intercept of the regression line projected back to zero-time. This is an estimate of the total amount of PBI which has been stimulated by the thyrotrophin per 100 ml. of plasma, or per 100 ml. of the distribution volume of thyroxine.

By the use of constant *a* (Characteristic 3), an estimate of the total amount of thyroxine which may be stimulated by a single injection of thyrotrophin (Characteristic 4) may be calculated. In the next section, it was estimated that the mean L-thyroxine space or volume of distribution was equivalent to 9.07% of body wt. Since PBI is nearly all thyroxine iodine, the total amount of thyroxine stimulated by the thyrotrophin may be estimated by the formula:

$$\text{total thyroxine stimulated, } \mu\text{g.} = \frac{a/0.653 \times \text{V.D.}}{100}$$

where a = intercept of PBI response curve on *Y*-axis at zero-time, PBI, μg. %
0.653 = fraction of thyroxine which is iodine by weight
V.D. = volume of distribution of L-thyroxine in ml. = body wt., g. × 0.0907

From Table 1 (Characteristic 4), the estimated total amounts of thyroxine stimulated by thyrotrophin ranged from 1.604 to 8.596 mg. Although the correspondence between total amounts of thyroxine stimulated and the doses of thyrotrophin injected was not good in the four trials, it did provide some useful estimates.

The mean slope value of rate constant *b* (Characteristic 5, Table 1) for the decline in PBI with time in the four trials was 0.0346; that is, a mean decline of 3.46% per hour in the PBI levels on an instantaneous basis. The slope or rate constants *(b)* may be more easily interpreted when presented as biological half-life values $t_{1/2}$ (Characteristic 6, Table 1), which are calculated by dividing 0.693 (natural logarithm of 2) by *b*, the slope or rate constant value. The biological half-life for PBI stimulated by thyrotrophin ranged from 14.3 to 28.9 hr., with a mean of 22.1 hr., which is similar to the mean biological half-life of 18.0 hr. obtained for PBI from exogenous L-thyroxine (next section and Table 2, disposal curve). O'Neal and Heinbecker *(10)* obtained a mean biological half-life of 16.2 hr. for the decline in PBI after the injection of a single dose of thyrotrophin in hypophysectomized dogs.

TABLE 2

Distribution and disposal characteristics of L-*thyroxine injected intravenously in dairy bull calves as measured by plasma PBI*

	H639[a]	G200[a]	G200[a]	Means
Age *(mo.)*	7.4	6.2	7.6	
Body weight *(kg.)*	264	233	198	
Normal PBI *(μg. %)*	3.47	4.29	4.07	3.94
Dosage hormone *(mg.)*				
Actual	2.86	2.86	5.72	
L-thyroxine equivalent	2.50	2.50	5.00	
Distribution curve (B)				
Degrees of freedom $(n-2)$	6	5	3	4.5
r[a]	−0.91[b]	−0.95[b]	−0.99[b]	−0.95[b]
a_1 *(PBI-μg. %)*	4.71	4.25	18.88	
$-b_1$ *(per hour)*	0.302	0.262	0.806	0.457
$t_{1/2}$ *(hr.)*	2.29	2.65	0.86	1.93
Disposal curve D				
Degrees of freedom $(n-2)$	6	6	10	7.3
r[a]	−0.98[b]	−0.98[b]	−0.98[b]	−0.98[b]
a_2 *(PBI-μg. %)*	6.57	8.63	17.07	
$-b_2'$ *(per hour)*	0.0406	0.0417	0.0343	0.0389
$-b_2^2$ *(per day)*	0.9744	1.0008	0.8232	0.9336
$t_{1/2}$ *(hr.)*	17.1	16.6	20.2	18.0
Volume of distribution				
Liters	24.9	18.9	19.1	
% Body weight	9.42	8.13	9.66	9.07
Thyroxine secretion per day				
Total *(mg.)*	1.285	1.242	0.995	
Per 100 lb. body wt. *(mg.)*	0.222	0.243	0.229	0.231

[a] Coefficient of correlation between time in hours after injection of thyroxine *(X)* and log$_e$ of PBI concentration, μg. %, *(Y)* of the first-order regressions.

[b] Significant at the 1% level ($P < 0.01$).

TABLE 3

Distribution and disposal characteristics of L-*triiodothyronine injected intravenously in dairy bull calves as measured by plasma PBI*

Characteristic	H639[a]	H639[a]	G200[a]	Means
Age *(mo.)*	6.2	7.3	7.8	
Body weight *(kg.)*	232	255	216	
Normal PBI *(μg. %)*	3.84	4.39	4.79	4.34
Dosage hormone *(mg.)*				
Actual	5.777	11.554	11.554	
L-triiodothyronine equivalent	5.580	11.160	11.160	
Distribution curve (B)				
Degrees of freedom $(n-2)$	1	5	6	4.0
r[a]	−0.999[b]	−0.98[b]	−0.99[b]	−0.99[b]
a_1 *(PBI-μg. %)*	8.48	16.82	18.50	
$-b_1$ *(per hour)*	1.92	2.47	2.42	2.27
$t_{1/2}$ *(hr.)*	0.36	0.28	0.29	0.31
Disposal curve (D)				
Degrees of freedom $(n-2)$	3	10	7	6.7
r[a]	−0.59[b]	−0.93[b]	−0.96[b]	−0.96[b]
a_2 *(PBI, μg. %)*	3.97	7.13	7.35	
$-b_2$ *(per hour)*	0.135	0.134	0.139	0.136
$t_{1/2}$ *(hr.)*	5.13	5.17	4.99	5.10
Volume of distribution				
Liters	82.3	91.7	88.9	
% Body weight	35.5	36.0	41.1	37.5

[a] Coefficient of correlation between time in hours after injection of triiodothyronine *(X)* and log$_e$ of PBI concentration, μg. %, *(Y)* of the first-order regressions.

[b] Significant at the 1% level ($P < 0.01$).

L-thyroxine and L-triiodothyronine. In a series of trials, the two bull calves were injected intravenously with sodium L-thyroxine pentahydrate (Table 2) and sodium L-triiodothyronine (Table 3) in varying quantities. Blood samples were collected at intervals over a period of approximately 72 hr. after injection in the L-thyroxine series and 12 hr. in the L-triiodothyronine series, and were assayed for PBI.

In both series of experiments, the levels of PBI declined rapidly during the initial phase following the injection of the materials and less rapidly thereafter. The methods of O'Neal *(9)* for the analysis of composite curves were applied to the various groups of experimental PBI-time series data.

The curves were each shown to have two components, each of which may be expressed as a first-order regression with an exponential rate of decline in which $\log_e$ of PBI concentration in the plasma was rectilinearly and inversely related to the time after the injection of the materials (Figure 2). The initial rapid decline in PBI, as represented by the initial first-order regressions (B curves, Figure 2), may be due to the distribution of thyroxine from the plasma into a larger body fluid compartment or space (volume of distribution). The slower decline in PBI concentrations, as represented by the second first-order regressions (D curves, Figure 2), may be due to the peripheral metabolism or disposal of the materials after distribution has been completed. Figure 2 presents representative trials for L-thyroxine and L-triiodothyronine on G200[6], in which the two first-order regressions are presented in each instance. The biological half-

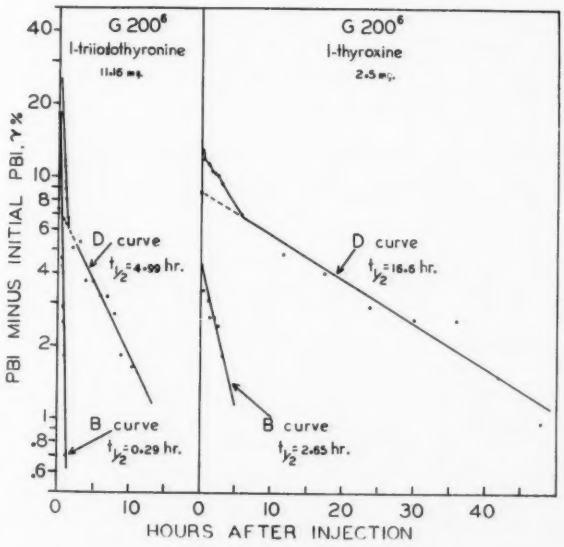

FIG. 2. Analysis of the curves of disappearance of PBI from the blood after the rapid injection of L-triiodothyronine (11.16 mg.) and L-thyroxine (2.5 mg.) in a Guernsey bull calf.

times ($t_{1/2}$) for distribution curves B were 0.29 and 2.65 hr. for L-triiodothyronine and L-thyroxine, respectively; whereas, the corresponding half-lives ($t_{1/2}$) for the disposal curves D were 4.99 and 16.6 hr., respectively.

The mathematical model for the disappearance of PBI from plasma is as follows:

$$\text{PBI, } \mu\text{g. \%} = a_1e^{-b_1t} + a_2e^{-b_2t}$$

in which a_1 = concentration of PBI (μg. %) at zero-time by extrapolation of the more rapid initial distribution curve,

$-b_1$ = slope (rate constant) of the distribution B curve, per hour,

a_2 = concentration of PBI (μg. %) at zero-time by extrapolation of the slower disposal D curve,

$-b_2$ = slope (rate constant) of the disposal D curve, per hour,

e = base of natural logarithms,

t = time in hours from injection of materials.

The biological half-life ($t_{1/2}$) for either of the rate reactions is calculated by dividing 0.693, the natural logarithm of 2, either by b_1 or by b_2, the rate constants.

Since a_2 equals the theoretical concentration of PBI in its volume of distribution at zero-time before any disposal has taken place, the volume of distribution (V.D.) of either thyroxine or triiodothyronine may be calculated by the following formula:

$$\text{V.D., ml.} = \frac{\text{dose } (\mu\text{g.}) \text{ of material} \times 100}{a_2/k_1 \text{ or } k_2}$$

in which k_1 = fraction of thyroxine (0.693) which is iodine by wt.

and k_2 = fraction of triiodothyronine (0.586) which is iodine by wt.

A summary of the kinetic characteristics for the distribution and disposal of L-thyroxine (Table 2) and L-triiodothyronine (Table 3) is presented for the two animals in the series of trials. In all instances, the coefficients of correlation *(r)* between the time in hours after injection of materials and the $\log_e$ of PBI of the several first-order regressions were high (range, −0.91 to −0.99), indicating that the mathematical model and the chemical analysis for PBI were satisfactory.

The rapidity with which L-thyroxine moved into its volume of distribution (mean distribution half-time, $t_{1/2}$, of 1.93 hr., Table 2) was much slower than the corresponding time for L-triiodothyronine (mean distribution half-time, $t_{1/2}$, of 0.31 hr., Table 3). O'Neal *(9)* reported a distribution half-time for D, L-thyroxine in dogs of 1.29 hr.

The mean disposal half-life for L-thyroxine ($t_{1/2}$, 18.0 hr., Table 2) was much longer than the mean disposal half-life for L-triiodothyronine ($t_{1/2}$, 5.1 hr., Table 3). The 22.1 hr. half-time of PBI stimulated by thyrotrophin (Table 1, Characteristic 6) was very similar to the 18.0 hr. disposal half-life for exogenous L-thyroxine (Table 2), which was an indication that the PBI in these instances

was primarily thyroxine iodine as contrasted to L-triiodothyronine iodine, which had a PBI disposal half-life of only 5.1 hr. O'Neal *(9)* found a disposal half-life of 24.6 hr. for D,L-thyroxine injected in dogs, and Recant and Riggs *(14)* reported a disposal half-life of PBI after injection of D,L-thyroxine in a normal human subject to be 4.3 days, or 103.2 hr.

The mean volumes of distribution for L-thyroxine (Table 2) and L-triiodothyronine (Table 3) were 9.07 and 37.5% of body wt., respectively. Triiodothyronine compared to thyroxine had a larger volume of distribution, faster rates of distribution and disposal, and lower iodine content, all reasons why L-triiodothyronine results in lower plasma levels of PBI in calves. O'Neal *(9)* reported that the volume of distribution of thyroxine in dogs was 36.9% of body wt., and Feldman *(4)* reported that the volumes of distribution for labeled thyroxine and triiodothyronine in the rat were 13 and 26%, respectively.

The method of Ingbar and Freinkel *(7)* was used to calculate the daily rate of thyroxine secretion (or degradation) of the two experimental animals (Table 2), using the data obtained in the three L-thyroxine infusion trials. These were calculated by multiplying the original PBI level by the volume of distribution of thyroxine by the $-b_2^2$ slope factor (fractional rate of thyroxine turnover), using the proper units in each instance, expressed on a daily basis. The daily rate of thyroxine turnover ($-b_2^2$) was obtained by multiplying the hourly rate of thyroxine turnover ($-b_2'$) by 24. The turnover values expressed as PBI were calculated in terms of thyroxine by dividing the completed calculations by 0.653, the fractional weight of iodine in thyroxine. The estimated amounts of thyroxine secreted (or degraded) by the animals per 100 lb. body wt. per day varied from 0.222 to 0.243 mg., with a mean of 0.231 mg. These trials were performed during the summer months (August and September), and the estimated daily thyroxine secretion rates agree well with those reported by Premachandra *et al. (11)* for cattle in summer. This is the first report presenting a practical method for determining thyroxine secretion rates in cattle which does not require the use of radioisotope facilities; therefore, it might have more extensive application.

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THE RATE OF MACHINE MILKING OF DAIRY COWS. II. EFFECT OF VACUUM AND PULSATION RATE[1]

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SUMMARY

Eighteen randomly selected slow-milking cows were milked with a machine operated at various vacuum levels (10, 12.5, and 15) and pulsation rates (20, 50, and 80) for a 27-day experimental period. The nine treatments were assigned at random at the start of each of the three replications included in the above experimental period. Increasing the vacuum level increased the speed of milking and the effect was greater from 10 to 12.5 than from 12.5 to 15 in. Increasing the pulsation rate increased the speed of milking, but the magnitude of its effect was much less than that of increasing the vacuum. Most of the effect of pulsation on increasing the speed of milking occurred from 20 to 50. Increasing the pulsation rate appeared to be more important at low levels of vacuum than it was at high-vacuum levels. Most of the effects of vacuum and pulsation on increasing milking rate were additive. A few treatment interaction terms were significant. In these cases the increases in milking rate expected, had the effects of vacuum and pulsation been additive, were not obtained as a result of this interaction. Changing the vacuum level or pulsation rate affected the cows differently. In general, increasing the vacuum or pulsation caused greater increases in milking rate, percentage-wise, for the slowest-milking group of cows than it did for the fastest-milking group of cows.

It was evident from the studies of Dodd *(4)*, Beck *et al. (2)*, and the authors *(8)* that a few cows substantially slow up the milking routine. If these cows could be milked faster, it would result in a great time-saving for most dairymen.

Smith and Petersen *(6)*, Baxter *et al. (1)*, and Gregoire *et al. (5)* obtained increased rates of milk flow upon increasing the vacuum level. Smith and Petersen noted considerable cow-to-cow variation in reaction to the increased vacuum. Baxter *et al. (1)* stated that the vacuum not only affected cows differently, but that increased vacuum caused greater increases in peak flow for fast-milking cows than for slow-milking cows. However, neither Baxter *et al. (1)* nor Whittleston and Verrall *(10)* obtained a decrease in over-all milking rate as they increased the vacuum, because longer stripping times were required.

Whittleston *(9)* observed that pulsation action was in some cases necessary for stimulation of milk flow, but when the pulsator was stopped after a normal start, the cows continued to milk at a normal rate in most cases. Whittleston and Verrall *(10)* found no difference in average rate of milking using 21 and 42 or 42 and 84 pulsations per minute and 14¾ in. of vacuum. On the other hand, Clough *et al. (3)* obtained faster milking with each increase in pulsation rate between 20 and 80.

None of these experiments was specifically directed at the slow-milking cow problem, nor was any designed to determine the interaction, if any, between

Received for publication July 18, 1957.

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vacuum level and pulsation rate. Furthermore, the effects reported on increasing vacuum level and pulsation rate were somewhat inconsistent. The purpose of this paper is to report the results of an investigation which was designed to clarify and add to our knowledge on this subject. Part of these data has been presented previously in abstract form *(7)*.

EXPERIMENTAL PROCEDURE

A standard single-unit, Sterling-type De Laval milker suspended from a spring-type scale was used throughout this study. The scale had been fitted into the frame of a small wooden stand which could be placed between the cows at milking time.

A small weight-controlled vacuum pump supplied the vacuum for the machine. The three vacuum levels used, 10, 12.5, and 15 in. of mercury, were obtained by changing the weights in the vacuum pump. The three pulsation rates used, 20, 50, and 80 per minute, were obtained by setting the adjustable Sterling-type pulsator before each milking.

All cows were milked at their regular milking time. Each udder was cleaned and massaged with an individual towel dipped in warm chlorine solution. This was immediately followed by the use of the strip cup. The machine was applied as soon as let-down had occurred. When the last cup was in place, the stop watch was started. Scale readings were taken at 15-sec. intervals until the milk flow fell to 0.3 lb. (0.2 for one cow) or less between readings. At this point, machine-stripping was started and continued until the cessation of flow; whereupon, the stop watch was stopped and the machine removed.

Eighteen slow-milking cows, which included two Brown Swiss, one Guernsey, and 15 Holsteins, were selected randomly for this experiment. Of these, six were milked during the winter, seven during the spring, and five during the summer months. Each of the cows required a longer total milking time than the average cow of its respective breed at the same stage of lactation. Each cow was milked for a three-day preliminary period before the 27-day experimental period started. This allowed the cows a chance to become accustomed to the special equipment and attention, and gave the operator a chance to become acquainted with the individual cow.

Before the start of the experiment, the cows were milked with a standard pail-type De Laval milker operated at 12.5 in. of vacuum and 48 pulsations per minute. During the preliminary period and the experimental period they were milked once a day with a machine operated as above and once a day with the experimental apparatus.

The rates of milking measurements used were as follows: *(1)* machine time (time to start of machine stripping), *(2)* stripping time, *(3)* stripping yield, *(4)* total milking time, *(5)* total yield, *(6)* average rate of flow, *(7)* maximum rate of flow (fastest minute), and *(8)* time to maximum rate of flow.

The design of the experiments is outlined (Table 1). It consisted of a 3×3 factorial arrangement of treatments. The resulting nine treatments were ran-

TABLE 1
Design of experiment[a]

Vacuum levels	Pulsation rates		
	20	50	80
10	11[b]	21	31
12.5	12	22	32
15	13	23	33

[a] All cows were milked with the same treatment on a given day.
[b] Treatment code numbers.

domly assigned at the beginning of each of the three replications. This provided three milkings for each of the 18 cows with each treatment, or a total of 54 milkings with each of the nine treatments.

For the analysis of variance the average of the three replications was used. The data were then analyzed according to the variance component model, X_{ijk}[3] = $\mu + V_i + P_j + C_k + VP_{ij} + VC_{ik} + PC_{jk} + VPC_{ijk}$, in which V and P were considered fixed and C was considered a random variable (V = vacuum, P = pulsation rate, C = cow). Individual degrees of freedom were assigned to V and P, to determine if the differences between treatment means were linear, quadratic, or both. Individual degrees of freedom were also assigned to the various components of between-treatment interaction, VP.

RESULTS

Statistical analyses were carried out on all the rate-of-milking measurements. The treatment, individual vacuum level, and pulsation rate, and experimental averages for all the rate-of-milking measurements are given (Table 2).

Effect of vacuum level. The right-hand column (Table 2) contains the average values obtained for each vacuum level. Machine time decreased from 6 min., 10 sec. at 10 in., to 5 min., 9 sec. at 12.5 in., to 4 min., 34 sec. at 15 in. The decrease from 6 min., 10 sec. to 4 min., 34 sec. was highly significant, linearly. Further study of the individual cow data showed that the slowest-milking group of cows was affected the most by the vacuum increase, which suggests that their stronger sphincter muscles played a role in determining milking rate. This observation is supported by the work of Baxter *et al. (1)*. They found that inserting cannulas of constant bore into the ends of the teats just prior to applying the machine caused the resultant maximum rate of milk ejection to be similar for all cows.

Stripping time decreased from 67 to 53 sec. as the vacuum increased from 10 to 12.5 in. No additional decrease was obtained at 15 in., however. This plateauing effect caused a significant quadratic trend. The amount of milk from machine-stripping was not significantly affected. Total milking-time decreased significantly. This trend was both linear and quadratic.

Total yield was found to increase linearly as the vacuum increased. This

[3] X_{ijk} is the observation on the k^{th} animal at the j^{th} pulsation rate and the i^{th} vacuum level.

TABLE 2

Effect of different vacuum levels and pulsation rates on milking rate: Average rates for 18 slow-milking cows. Each cow was milked three times with each treatment

	Vacuum levels	Pulsation rates			
		20	50	80	x
Machine time (*min. and sec.*)	10	6:49	5:58	5:42	6:10
	12.5	5:31	5:04	4:53	5:09
	15	4:54	4:26	4:23	4:34
	x	5:45	5:09	4:59	5:18
Significant: V_l^a, P_l^a, P_q^a, $V_l \times P_l^a$					
Stripping time (*sec.*)	10	75	59	67	67
	12.5	51	54	55	53
	15	58	50	55	54
	x	62	54	59	58
Significant: V_l^b, V_q^b					
Stripping yield (*lb.*)	10	1.98	1.76	2.05	1.93
	12.5	1.70	2.03	2.10	1.94
	15	2.41	2.17	2.44	2.34
	x	2.03	1.99	2.20	2.07
Significant: None					
Total milking time (*min. and sec.*)	10	8:04	6:57	6:50	7:17
	12.5	6:22	5:57	5:48	6:03
	15	5:50	5:16	5:18	5:28
	x	6:45	6:03	5:59	6:16
Significant: V_l^a, V_q^a, P_l^a, P_q^a, $V_l \times P_l^a$					
Total yield (*lb.*)	10	20.4	19.5	20.2	20.0
	12.5	20.7	20.9	20.4	20.7
	15	21.7	21.4	20.9	21.3
	x	20.9	20.6	20.5	20.7
Significant: V_l^a, P_l^b, $V_l \times P_q^b$					
Average rates of flow (*lb/min*)	10	2.6	2.9	3.1	2.9
	12.5	3.3	2.6	3.6	3.5
	15	3.8	4.1	4.0	4.0
	x	3.3	3.5	3.6	3.4
Significant: V_l^a, V_q^b, P_l^a, P_q^a, $V_l \times P_l^b$					
Maximum rate of flow (*lb/min*)	10	3.9	4.1	4.4	4.1
	12.5	4.8	5.3	5.3	5.2
	15	5.5	6.0	6.0	5.9
	x	4.7	5.1	5.2	5.0
Significant: V_l^a, V_q^b, P_l^a, P_q^b, $V_l \times P_q^b$					
Time to maximum rate of flow (*sec.*)	10	152	107	112	124
	12.5	103	91	83	93
	15	84	66	67	72
	x	113	88	88	96
Significant: V_l^a, V_q^a, P_l^a, P_q^b					

V—Vacuum; P—Pulsation; l—linear; q—quadratic.
[a]Significant at 1% level.
[b]Significant at 5% level.

may have been owing to the slow milking, which tended to allow the effect of oxytocin to wear off before the cows were completely milked.

Maximum rate of flow increased from 4.2 at 10 in. to 5.2 at 12.5 in. to 5.8 at 15 in. This trend also was both linear and quadratic.

Effect of pulsation rate. Average values obtained for each pulsation rate are shown (Table 2). In general, increasing the pulsation rate significantly increased the speed of milking, but not as much as did increasing the vacuum

level. This effect of increasing pulsation rate was most marked on increasing it from 20 to 50. No noticeable effect occurred on increasing the rate from 50 to 80. This means that 20 pulsations per minute was definitely too slow for all the vacuum levels used. This fact is emphasized by Figure 1. In this figure, the average total milking time of each treatment is represented by a dot. It

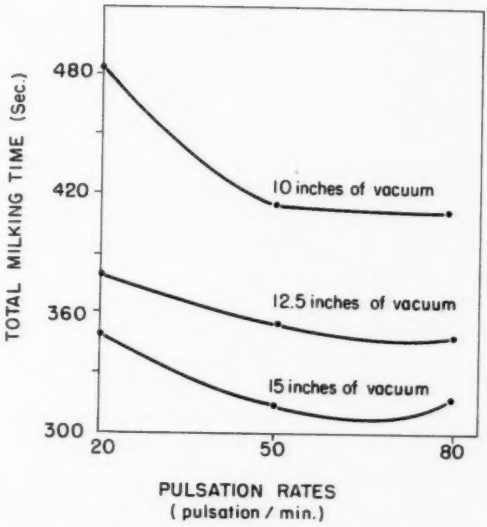

FIG. 1. Effect of different vacuum levels and pulsation rates on total milking time.

can be seen that increasing the pulsation rate from 20 to 50 was more important at the low level of vacuum than at higher ones.

Effect of interaction between different vacuum levels and pulsation rates. By assigning individual degrees of freedom to the various components of between-treatment interaction, it was possible to determine which components were important. These are enumerated (Table 2). Actually, few were statistically significant, indicating that the effects of vacuum and pulsation on milking rate are primarily additive.

The vacuum linear by pulsation linear component, $V_1 \times P_1$, was highly significant for machine-time and total milking time, and significant, $P < .05$, for average rate of flow. In these cases, the significance occurred because the combination of vacuum and pulsation resulted in slower milking than expected, based on the additive effects of vacuum and pulsation.

Effect of vacuum and pulsation on different cows. Great variation was found between the 18 slow-milking cows in their rate of milking. In fact, the magnitude of the between-cow mean squares indicated that this was the major source of variation in most of the analyses.

The various levels of vacuum affected the individual cows differently. This conclusion is based on the fact that the vacuum by cow interaction, VC, was highly significant for all the rate-of-milking measurements except stripping time, which was significant ($P < .05$). The three pulsation rates also affected the cows differently, since the pulsation-by-cow interaction, PC, was highly significant for total milking time, average rate of flow, and maximum rate of flow.

DISCUSSION

As was found by Smith and Petersen *(6)*, Baxter *et al. (1)*, and Gregoire *et al. (5)*, this study showed that as the vacuum increased the maximum rate of flow increased. As to magnitude of increase, Smith and Petersen *(6)* obtained a 100% increase in peak flow (maximum rate of flow) upon increasing the vacuum from 10 to 15 in. In this study, the maximum rate increased by only 40.5% as the vacuum increased from 10 to 15 in. Smith and Petersen found that more of the increase occurred between 10 and 12.5 than between 12.5 and 15, which is what occurred in this study.

Baxter *et al. (1)* reported that they failed to obtain an increase in average rate of flow as they increased the vacuum from 11 to 20 in., owing to the longer time necessary to remove the large amounts of strippings left at the higher vacuum levels. Whittleston and Verrall *(10)* also obtained no significant differences in average rate of flow between 10 and 14¾ or between 14¾ and 19 in. of vacuum. However, in our study, the average rate increased at approximately the same rate as the maximum rate of flow, 37.8, as compared to 40.5%.

Smith and Petersen *(6)* secured a lower amount of machine strippings at 14 in. of vacuum than at 10, 12, or 16 in. of vacuum, and Gregoire *et al. (5)* obtained smaller amounts of machine-strippings at 13 than at 10 or 17 in. of vacuum. In this study, there were no significant differences in amount of strippings between the three vacuum levels used, although the largest amount of strippings was obtained at 15 in. of vacuum. Gregoire *et al. (5)* also reported no significant differences in stripping time; whereas, in this experiment, the stripping-time decreased as the vacuum increased from 10 to 12.5. This is attributed to the fact that more pull could be applied to the teat cups while machine-stripping as the vacuum was increased. However, as the vacuum increased from 12.5 to 15, the stripping-time tended to remain about the same because of the somewhat larger amounts of strippings obtained at 15 in.

Gregoire *et al. (5)* found no significant difference in total yield over a 40-wk. period between 10, 13, and 17 in. of vacuum on 12 first-calf heifers. The 18 slow-milking cows in this study, however, showed a highly significant increase in total yield as the vacuum increased. Apparently, at the lower vacuum levels the effects of the oxytocin had begun to wear off because of the slow milking, before some of the cows were completed.

Similar to the findings of Smith and Petersen *(6)* and Baxter *et al. (1)*, great cow-to-cow variation was noted in the reaction of the cows to the various vacuum levels. Baxter *et al. (1)* found that the two fastest-milking cows had a

greater increase in maximum rate of flow, as the vacuum increased, than did the two slowest-milking cows in their study. A similar phenomena was noted in this study; although, percentage-wise, the slow-milking cows increased much more than did the faster-milking cows. The average and maximum rates of flow are compared for the four fastest- and four slowest-milking cows (see Table 3).

TABLE 3

Comparison of the average and maximum rates of flow for the four slowest and the four fastest cows at different vacuum levels and pulsation rates

	Average rate *(lb/min)*		Maximum rate *(lb/min)*	
Vacuum level	4 Slowest cows	4 Fastest cows	4 Slowest cows	4 Fastest cows
10	1.53	4.26	2.04	6.60
12.5	2.12	4.85	2.77	7.52
15	2.50	5.28	3.26	8.62
Diff. 10–12.5 inc.	0.59	0.59	0.73	0.92
	38.5%	13.9%	36.0%	14.0%
Diff. 10–15 inc.	0.96	1.02	1.23	2.02
	62.7%	24.0%	60.3%	30.6%

	Average rate *(lb/min)*		Maximum rate *(lb/min)*	
Pulsation rate	4 Slowest cows	4 Fastest cows	4 Slowest cows	4 Fastest cows
20	1.91	4.55	2.48	7.45
50	2.12	4.90	2.81	7.62
80	2.11	4.94	2.78	7.68
Diff. 20–50 inc.	0.21	0.36	0.33	0.17
	11%	8%	13%	2%
Diff. 20–80 inc.	0.19	0.39	0.30	0.23
	10%	9%	12%	3%

The magnitude of the increase in average rate is about the same for both groups but, percentage-wise, the increase is much greater for the slower-milking cows.

Whittleston and Verrall *(10)* noted that the teat cups tended to fall off more when 10 in. was used than when higher vacuums were used. A similar observation was noted in this study.

Whittleston and Verrall *(10)* found no statistically significant increase in milking rate upon increasing the pulsation rate from 21 to 42 or from 42 to 84 with the vacuum at 14¾ in. However, Clough *et al. (3)* obtained an increase in milking rate on increasing the pulsation from 20 to 80 at 15 in. of vacuum. Increasing the pulsation from 20 to 80 also resulted in faster milking in this study. Clough obtained a 1.39 lb.-per-minute increase in peak flow and a 0.34 lb.-per-minute increase in average rate of flow upon increasing the pulsation from 20 to 80. In this experiment, the maximum rate increased by 0.50 lb. per minute and the average rate of flow by 0.31 lb. per minute. Furthermore, they obtained a 1.26-min. decrease in total milking time as compared to 0.78 min. in this experiment. Actually, in this experiment no apparent increase in milking rate was obtained by increasing the pulsation from 50 to 80. Apparently, as the vacuum increased, the effect of an increase in pulsation rate was of progressively

less importance in increasing the rate of milking. However, 20 pulsations per minute appears below the optimum rate, even at 15 in. of vacuum.

Pulsation also affected the cows differently (shown in Table 3). The slowest-milking cows had about the same percentage increase in average rate of flow as did the four fastest-milking cows, but the slow group had a greater increase in maximum rate of flow than did the fast group as the pulsation increased.

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TECHNICAL NOTES

SOME VOLATILE COMPONENTS OF CHEDDAR CHEESE[1]

The chemical character of Cheddar cheese aroma has been under investigation at this and other laboratories for a number of years *(3, 4)*. Regarding the relative importance of any given compound or class of compounds in the aroma (e.g., fatty acids, carbonyls, amines, etc.), there seem to be substantial differences of opinion, not only between investigators but among testers. One may conclude that Cheddar aroma means different things to different people and that the total aroma is a complex mixture. Some compounds which we have identified as components of this mixture are dimethyl sulfide, ethanol, acetone, diacetyl, butanone-2, and 3-hydroxybutanone. Presumptive evidence of heptanone-2 also has been secured. With the possible exception of the latter and diacetyl, only dimethylsulfide, which was observed to be a "top-note" in all good quality Cheddar, is considered to be of obvious and direct importance to the aroma mixture.

Experiments with gas chromatography. Sections from full-cream Cheddar cheeses of good quality ranging in age from 6 mo. to a year were freed of rind and ground in a meat grinder. The ground cheese, in amounts of 1 to 5 kg., was mixed with an equal weight of distilled water. The volatiles in this mixture were removed, trapped, and analyzed by gas chromatography (GC) and mass spectrometry, using the equipment and procedures as described by Day *et al. (1)*. The distillation was carried out at from 40 to 50° C. and 30 to 40 mm. Hg pressure. Only the components recovered in dry ice–ethanol and liquid nitrogen traps were analyzed. On the basis of retention volumes by GC and mass spectral analysis of the components thus separated, dimethyl sulfide, ethanol, acetone, and diacetyl were conclusively identified. The wet-ice trap invariably had an odor suggestive of heptanone-2. A small GC peak corresponding in retention volume to that of butanone-2 also was observed. Further evidence of these two ketones in the volatiles is presented below. Several unidentified GC components with ester-like odors were noted. Although quantities of volatile components, as indicated by peak areas, appeared to vary between lots of cheese, the same components were noted rather consistently.

Experiments with volatile carbonyls: One-half kilogram quantities of ground cheese, as described above, were mixed with one liter of distilled water. This mixture was steam-distilled at atmospheric pressure. Only the first 250 ml. of distillate was collected and this was led directly from the condenser into a solution of 100 mg. of 2,4-dinitrophenyl (DNP) hydrazine dissolved in 1 ml. of concentrated H_2SO_4 and 2 ml. of water. After standing one hour, this solution was extracted with 50 ml. of benzene to recover the DNP hydrazones. The benzene extract was evaporated to a volume of 5 ml. and a 5-λ quantity of this solution paper-chromatographed by the method of Huelin *(2)*. Analysis of University cheese, as well as of six retail store samples of Cheddar, revealed DNP hydrazones falling into six well-defined groups, based on R_f values. These are shown (Table 1), together with some data on their identity. It is to be emphasized that the spots observed may have arisen from more than one compound. All the cheese samples showed evidence of most if not all of the R_f components. However, the spot at R_f 0.74 was always faint and sometimes absent. All samples showed immobile spots at the base line of chromatograms. It was assumed that these resulted at least in part from reagent and the bis-DNP hydrazone of diacetyl.

[1] Authorized for publication on February 28, 1958 as paper No. 2238 in the Journal Series of the Pennsylvania Agricultural Experiment Station.

TABLE 1

Some properties of 2,4-dinitrophenylhydrazones from volatile carbonyls of Cheddar cheese

R_f of unknowns [a]	Knowns of corresponding R_f [b]	Absorption maximum in ethanol *(mμ)*		M.P. (° C.)		
		Known	*Unknown*	*Known*	*Unknown*	*Mixed*
0.74	Heptanone-2	362	362	58, 72, 89		
0.61	Butanone-2	362	362	115	113	113-115
0.46	Acetone	362	361	127	.....	.....
0.29	Acetaldehyde	360	359	157, 168	.....	.....
0.19	Formaldehyde	348	350	166	.....	.....
0.06	3-OH butanone	357.5	357	115 [c]	112	112-115

[a] Paper chromatography by the method of Huelin *(2)*.

[b] Other DNP hydrazones have similar R_f's. Those derivatives listed either are considered plausible or are confirmed by data of this paper.

[c] Mono derivative.

The benzene extract of DNP hydrazones from several cheeses showing heavy spots at R_f 0.61 were pooled, evaporated to dryness, and extracted with petroleum ether (boiling, 38 to 42° C.). This extract was evaporated to dryness and the residue was recrystallized repeatedly from ethanol, yielding a maximum constant m.p. of 113° C. Admixture with an authentic sample of the butanone derivative showed no depression (Table 1).

The benzene extract of DNP hydrazones from a cheese showing an intense spot at R_f 0.06 was evaporated almost to dryness and the remaining solution streaked along the entire baseline of several chromatograms. Following development, the areas at R_f 0.06 were cut from the papers, eluted with ethanol, and the ethanol extracts evaporated nearly to dryness. Crystallization was induced by adding water. The crystals were recovered, dried, and recrystallized from a mixture of benzene and petroleum ether. This product, m.p. 112°, could not be recrystallized further, owing to limitations of quantity. It showed no depression in mixed melting point with an authentic derivative of 3-hydroxybutanone and showed essentially the same absorption maximum (Table 1).

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OBSERVATIONS ON THE DIGESTIBILITY OF ORCHARDGRASS PASTURE AS AFFECTED BY SEASON AND GRAZING MANAGEMENT[1, 2]

Recent developments *(1, 2, 4)* in research methods have made possible the determination of the grazing animal's dry matter consumption and digestibility of forages. The Western Washington Experiment Station, Puyallup, is currently using these techniques to ascertain the relative nutritive value of various grass and legume species. Some preliminary observations from this work may interest others working in this area.

Reid *(3)* previously reported a rapid decline in dry matter digestibility of first-cutting forage with advancing maturity, after a base date of April 30. Dry matter digestibility declined from approximately 77% in early May to approximately 52% by mid-July. A similar decline in digestibility with advancing maturity was reported for aftermath forage, but the initial digestibility values were not so high and the decline was not so rapid as with first growth. The digestibility of the aftermath was approximately 65% at 20 days after first cutting and declined to approximately 57.5% at 70 days.

Swift *et al. (5)* determined the digestibility of successive cuttings of orchardgrass by sheep. The forages were dried immediately after cutting. A seasonal decline was noted from spring to fall cuttings. Adding supplementary nitrogen immediately after the third cutting increased the nitrogen content of the succeeding cutting and increased the apparent digestibility of the dry matter.

The orchardgrass (*Dactylis glomerata* var. *Akaroa*) pasture used in this study was a relatively pure stand. The pasture area of approximately one acre was divided into three equal plots for rotational grazing. Three yearling Holstein heifers were put to pasture on these plots beginning April 18 and pastured continuously throughout the summer.

Grab samples of feces were collected at 6 A.M. and 4 P.M. for chromogen analyses, begin-

[1] This study was supported in part by funds from the Dairy Husbandry Research Branch under Cooperative Agreement 12-14-100-769(52).

[2] Scientific Paper No. 1693, Washington Agricultural Experiment Stations, Pullman. Project No. 1333.

ning 12 days after going on pasture. The A.M. and P.M. fecal samples were combined on an equal-weight basis for a period of seven days.

Dry matter digestibility was determined from the chromogen analyses of the feces samples, according to the method of Reid *et al. (4)*. Two digestion trials were conducted during the 1957 pasture season. The fecal collection periods during the first trial began on April 30 and continued through May 6; whereas, the second trial began on July 5 and continued through July 11. A series of five digestion trials throughout the pasture season had been planned, but technical difficulties limited the number of trials which was possible.

The dry matter digestibilities of orchardgrass by three dairy heifers during the two digestion trials are shown by days (Table 1). The individual determinations are presented to show the consistency of the daily trends between animals.

TABLE 1

Daily dry matter digestibility of orchardgrass pasture by Holstein heifers

Date	Animal No. 591	593	594	Average
		(% digestibility)		
	Trial 1			
April 30	73.8	72.6	72.8	73.1
May 1	73.6	72.4	72.1	72.7
May 2	71.9	71.7	70.2	71.3
May 3	71.5	71.2	69.4	70.7
May 4	69.6	70.9	68.8	69.8
May 5	69.0	69.1	67.5	68.5
May 6	66.9	68.0	66.9	67.3
		7-day average		70.5
	Trial 2			
July 5	74.2	73.8	72.8	73.6
July 6	72.9	73.5	72.6	73.0
July 7	71.8	73.6	71.6	72.3
July 8	68.8	72.3	69.3	70.1
July 9	67.0	70.7	69.5	69.1
July 10	71.9	71.1	72.2	71.7
July 11	73.7	75.4	74.1	74.4
		7-day average		72.0

The orchardgrass was in the early boot stage when fecal collections were started during Trial 1 and in the vegetative state during Trial 2.

Advancing maturity and grazing behavior may account for the daily variations observed in digestibility. The heifers were pastured continuously on one plot during the fecal collection period of Trial 1. The marked daily decline in digestibility during this trial partly may be a result of advancing maturity and partly due to selective grazing. Under rotational grazing the animals tend to eat the more leafy and digestible portions of the plant first when admitted to a new pasture plot. Consequently, on successive days the grazed forage becomes more stemmy and less digestible.

During Trial 2, the heifers were rotated to a new pasture plot on July 9. Therefore, the first five days of this trial represented forage that had been selectively grazed for a period before the start of the trial; whereas, the last two days represented the choice forage selected from fresh regrowth. A decline in digestibility during the first five days was followed by an increase in digestibility during the last two days. Differences in stage of maturity of the aftermath of these two plots were small at this season of the year.

These results indicate some possible errors in pasture digestibility measurements with animals on a rotational system of grazing. The fecal sampling periods should be comparable with the length of the grazing period for the entire plot. Short sampling periods, either at the beginning or toward the end of a grazing period, may not be indicative of the digestibility of the entire plot. The use of strip-grazing may be advantageous for such studies because the quality of grazed forage is more uniform.

There was no apparent decline in digestibility from early spring to aftermath forage, as reported by investigators previously. Inasmuch as orchardgrass tends toward early maturity on first growth, the forage grazed during Trial 1 may have been slightly more mature than that during Trial 2. The orchardgrass plots were maintained at a fairly high level of fertility throughout the season by the application of approximately 35 lb. of available nitrogen per acre after each grazing. Also, the plots were irrigated adequately during the grazing season.

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OUR INDUSTRY TODAY

A STUDY OF MILK DATING IN THE CITY OF NEW YORK[1]

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This study of the dating of milk in New York City was conducted during April and May, 1957. Historical and general material is limited to that necessary for a clear presentation of the results. Complete data will be available in a bulletin from the Cornell University Agricultural Experiment Station.

About 45 yr. ago, the Board of Health of the City of New York first made it mandatory, as a public health measure, to mark on the retail milk container, or bottle, the day of the week when milk was pasturized. At that time, three grades of milk could be sold in New York City, and only Grade A was suitable for children. There can be no doubt concerning the wisdom of dating milk as a public health measure in that period of the early development of a sanitary, wholesome milk supply.

Since those early days in food sanitation there have been great advances in the quality of milk and in its processing, distribution, and handling. Today the quality of milk in food stores can not be related to the day of pasteurization; consequently, it is questionable whether any public health reason remains to justify this dating regulation. Milk dealers object to this requirement as it increases the cost of milk distribution and dairy farmers suspect that it reduces sales of milk in their most profitable market. On the other hand, it is apparent that some boards of health in cities with the dating regulation are concerned with possible adverse public reactions to the removal of the regulation. At present, only 18 cities in the United States with a population over 100,000, and only two states, require dating. Last year, according to press reports, the boards of health in two of these 18 cities recommended against dating milk as a public health measure, but the common councils retained the provision. It is interesting to note that the laws of the State of New York make no provision for dating and the New York State Department of Health has opposed such a regulation within the state.

When some dairy farm organizations became concerned about this requirement in the New York City milkshed, an Assistant Commissioner in the City Department of Health pointed out that a study of the situation was necessary to ascertain the pertinent facts. At a meeting called by the New York State Commissioner of Agriculture and Markets to discuss this problem, he requested that a plan be made for such a study. Both the State Department of Agriculture and the City Department of Health offered personnel to conduct the study. Milk producers and dealers also offered to cooperate. The study was to be made immediately, to obtain enough data to show the effect of dating on milk sold through food stores. It was agreed that home deliveries and services in restaurants and hotels should not be included because dating could not be a milk-quality factor in these markets.

Areas within the city were selected on the basis of various levels of consumer income, and four types of stores were designated. Then, city sanitarians and state food inspectors were asked to select food stores of these types in the selected areas within their usual territories. These food stores were studied for one day. Also, a smaller number of food stores were studied for seven consecutive days. In adjacent municipalities outside New York City, stores were studied which handled milk marked by company code and with no time-limit on sale.

In addition to the food stores, the study included representative milk-processing plants within the City of New York, and one plant outside the city that code-marked all milk containers.

Within the city, the 187 food stores studied for one day handled 57,342 quart paper containers of milk daily, or 307 quarts per store. The wholesale deliveries were usually made before 9 A.M. and very few milk drivers rotated the dated milk in the refrigerated sales case. The 34 stores outside the city that were studied for one day handled 8,186 quarts of coded milk daily, or 241 quarts per store. Deliveries of coded milk were generally made after the store personnel arrived, and about one-third of the routemen rotated milk in the sales case.

Results with Stores Studied for One Day

The grocers' statements (and sometimes their records) showed that at least twice a month three out of four food stores had no milk for sale at closing time. The stores closed without milk an average of 1.6 times per week per store, or one-fourth of the time. One food store in eight generally had no milk for sale at closing time, and only one store in four generally had milk for sale all the time. In contrast to this prevalent lack of dated milk for sale in food stores at closing time, 67% of all grocers handling coded milk were seldom or never out of milk.

The most accurate check on the availability of milk for sale at all times was the presence or

[1] Presented at Eleventh Annual Meeting, Dairy Products Improvement Institute, Inc., February 13, 1958, New York, N. Y.

absence in the store "last night" at closing time, as reported to the survey workers. Data obtained on this basis showed that one-third of all stores handling dated milk closed business at the end of the day with no milk for sale. A Monday check revealed that 47% of all stores had been without milk when closed on Saturday or Sunday. A striking difference was found for coded milk: only one store out of 34 closed with no milk on hand.

The age of milk purchased by consumers can be ascertained most accurately and easily by an actual count of the quart paper containers offered for sale in the food stores. Milk dated today constituted 91% of the total milk on hand; 3.6% was yesterday's milk, and only 0.3% was two or three days' old, according to the date on the container. No older milk was found, even though 38,237 quarts were counted in this part of the study. An interesting aspect of this picture is that 4.5% of all milk in stores was tomorrow's milk. Predating 36 hr. is legal in the City of New York, but delivery of such milk in advance of the day marked on the container is illegal. To obtain the true age of the milk from the actual hour of pasteurization from 15 to 34 hr. must be added to the time interval.

The coded milk studied was sold outside of, but adjacent to, New York City without date and with no time-limit. Slightly over 82% of the coded milk was marked as today's milk, 15 % was yesterday's, and 2% was two or three days' old. The age of dated and coded milk as sold in food stores in New York City and in adjacent areas was the same, except for a shift of 10% from today's to yesterday's milk.

It was the custom of half of the stores to sell milk dated Saturday on Monday morning, since this time span was within the legal limits of the Sanitary Code, provided the sale was made before noon. Some stores returned some of the milk to the plants on Monday, to decrease the amount of Saturday's milk to be sold. Other stores returned all milk to the plants on Monday morning. On days not following Sundays or holidays. it was customary for three stores in four to sell yesterday's milk; that is, one store in four returned yesterday's milk to plants.

When a purchase of milk was made, the survey worker noted whether the customer examined the containers. Those who did were queried, without any leading questions, to learn why they looked at or examined the containers. All customers were classed about equally in three groups: those who did not examine containers, those who examined containers to see whether they were leaking milk, and those who looked for the date. It appears, therefore, that two-thirds of all consumers bought dated milk without looking for the date. More than half of all purchases of coded milk sold outside New York City were made without examining the container.

Results with Food Stores Studied for Seven Consecutive Days

The nine food stores handling dated milk in New York City which were studied for seven consecutive days received a total of 19,288 quart containers on regular deliveries and 1,000 quarts on special deliveries. The largest deliveries were received on Saturday and Monday. Each store closed yesterday's business with an average of 35 quarts of milk on hand, and on an average each store closed once a week with no milk for sale. Each store returned 54 quarts of yesterday's milk to the milk dealer, or 2% of all deliveries. About 90% of all milk in the stores was dated today, the remainder being equally divided between tomorrow's milk and yesterday's milk.

The four food stores handling coded milk (with no date and no time limit on sale) outside New York City, which were studied for seven consecutive days, received a total of 8,134 quarts on regular deliveries, and there were no special deliveries. The amount of milk delivered daily to each store was fairly uniform. Each of these stores closed yesterday's business with three times as much milk on hand as in the stores handling dated milk. Each store handling coded milk always had milk for sale at the close of every day's business. No coded milk was returned to processing plants for credit. About 84% of all milk in the stores was coded as today's milk; the remainder was yesterday's milk, except for 14 quarts of two-day-old milk in one store when it opened in the morning.

The dated and the coded milk were being sold within brief, comparable periods of time after pasteurization.

Studies in Milk Plants

The 11 pasteurization plants studied in New York City included the range of volume of business. They represented 31% of all plants and 28% of all milk processed for the entire city.

Packaging of milk marked with tomorrow's date began at any time between 8 A.M. and 8 P.M., according to company records. The time interval from the beginning of pasteurization to the time indicated in the Sanitary Code "for distribution after 6 A.M." of the following day varied from 10 to 22 hr. and averaged 15 hr. Actual observations by the survey workers showed this average time to be 19 hr. from pasteurization to date.

The Sanitary Code provides that the maximum interval for the date "shall not be more than thirty-six (36) hr. after pasteurization." This extra time is allowed for predating milk day-after-tomorrow to handle extra needs over holidays and weekends. The time interval from the beginning of pasteurization to date on predated milk varied from 21 to 43 hr. and averaged 34. Although predating is not universal, the plant records showed that a maximum of 50%

and an average per plant of 20%, of all processed milk was predated.

In contrast, it is well to point out that the number of hours from pasteurization to the time designated by the code for milk sold outside New York City was only 10 hr. Consequently, the age of coded milk as compared with dated milk may be slightly better than shown by the data.

Data prepared by the milk companies showed that 2% of all dated milk loaded on wholesale trucks was not sold and was returned to the plants, an amount ten times greater than for coded milk. The company data revealed that 0.6% of all dated milk sold to food stores was returned for credit, and the survey workers found the amount to be 0.7%. No coded milk (actually one quart) was returned by the retailers for credit, as found by the survey workers. These returns of dated milk were less than anticipated, but they do not include unsold today's milk (2% of the load) and unsold tomorrow's milk (3.2% of the load) returned to the plant. Together these represent 5.2% of the entire wholesale truck load of dated milk. Of the milk returned for credit from the stores to the plants, 0.41% of the total deliveries was yesterday's milk, 0.31% was two-day-old milk, and 0.06% was from three to seven days old. In view of the fact that milk beyond the three-day age was never found in the sales cases of food stores, it must be assumed that this small amount of old milk represented milk containers set aside until the truck routemen picked them up for credit.

Interviews with 85 wholesale routemen revealed that half of the deliveries of dated milk were made before the stores opened. Very few routemen rotated the milk for the grocers, even though one-fourth of them placed the milk in the sales case as instructed by the man in charge of milk at the food store. This study revealed that routemen underestimated dated milk returned to plants. Predated milk was handled on 78% of all routes and the routemen's estimates or figures showed it to be 21% of the total load, an amount in close agreement with company records.

About 4% of all processed dated milk was delivered wholesale by special deliveries, an item that must add to the cost of milk distribution. Special deliveries were a necessary part of distributing dated milk, but were held to almost nothing in the handling of coded milk.

Discussion of Study and Problem

Any discussion of sanitary milk regulations and milk quality must be predicated on the premise that safe milk of excellent quality shall be available to consumers at all times. The health department is concerned with its responsibility for safeguarding the health of its citizens, through its efforts to insure that only milk of fine quality is sold within the city. The dairy industry, particularly milk producers and milk dealers, depends for its very existence upon consumers receiving milk that is satisfactory in every respect, because consumers pleased with the quality of milk will make future purchases. The dairy industry and health departments have worked together closely in recent decades to give the citizens of this nation one of the world's finest milk supplies. The Department of Health of the City of New York, state and local regulatory agencies, educational agencies, equipment and supply companies, and the dairy industry of this milkshed have been active leaders in this progress.

The principal and only argument of consequence used to favor the dating of milk containers has been the assumption that consumers would receive milk of less age if dated and if a time limit were fixed for sale. Most persons familiar with the problem realize that the high quality of today's milk supply, together with technological and sanitation advances in the production, processing, and distribution of milk have reduced to insignificance any time supposedly gained through dating. Age of fresh milk is of much less importance than quality and handling procedures. The present study of the milk supply of New York City reduces such discussion to an absurdity, because the age of dated milk sold under a time-limit in food stores was the same as that of secretly coded milk sold with no time-limit, in neighboring municipalities.

The dating of milk is distinctly objectionable for the following reasons:

1. It places false emphasis on a date that has not affected the age of the milk and that may not reveal the age of the milk.

2. It misleads the public by associating a date with milk quality. Emphasis should be on quality only. The large majority of the cities (all but 18 over 100,000 population) and most states (all but two) do not have dated milk and there is no evidence to indicate any detrimental effect on milk quality of marketing nondated milk with no time-limit on sale.

3. It must reduce milk sales, as an appreciable percentage (34%) of food stores handling dated milk had no milk for sale at closing time. Food stores outside New York City which sold secretly coded milk almost always had milk on hand for sale.

4. The dating regulation is a needless distribution expense and irritation to the dairy industry. There is the loss owing to substantial amounts of milk being returned to plants for dumping, and the expense of special deliveries. There is the constant endeavor to exactly balance deliveries and sales in food stores, and to avoid sales of yesterday's milk to certain customers. These objectionable features of milk dating may be eliminated by coding.

Therefore, since the dating of milk containers serves little or no purpose today in sanitary milk distribution and public health control, it is recommended that dating should be deleted from the Sanitary Code. The regulations should require secret company codes with no change in the time limit from pasteurization to sale. Such a requirement will insure that no adverse change will occur in the age of the milk at any time, and that the present routine of milk deliveries will not be affected by the elimination of dating.

Some persons have expressed concern about the possibility that certain civic, social, or political leaders might arouse emotional reactions against the removal of the dating provision. In some instances, agitation by organized minority groups have produced ill will; and in others, such agitation was completely ignored by the public. I believe that any reasonable action of a board of health, taken after careful consideration of the problem, will be accepted in good spirit by an overwhelming majority of the citizens and their group leaders, because they have faith in the integrity and ability of the board members. The history of public health in this country has shown this to be true. Occasionally, criticism of active progressive leadership is voiced temporarily by minorities, and the certain approval of the majority may be delayed or not expressed. Some criticism of persons in high positions is to be expected, but it is not always forthcoming.

It has been said, too, that the public will not allow the dairy industry to save this needless expense in milk distribution, because it is not sufficient to warrant a reduction in the retail price of milk. I believe that this idea is not worthy of serious consideration in our public-spirited nation. When it is realized that the elimination of dating will not affect the age or quality of milk or the routine of distribution, but will save money in milk distribution costs and will increase sales, there will be no question about public acceptance.

An example of good acceptance of decisions made in the public interest was shown in a city that replaced dating by compulsory secret coding in 1955. In reply to my questions of January, 1957, the Senior Sanitarian in charge of milk sanitation in the city health department wrote comments which included the following statements:

"Some difficulty was experienced in selling the City Commission on coding and taking off the limited hours milk could be sold.

"One member of the City Commission worked very hard to have the amendment stopped or delayed, and especially to keep the 72-hr. limit on time the milk must be sold.

"We have had no trouble that I have heard of since the new amendment to the ordinance has been enforced. I certainly would hear of any troubles from using the dating code, if there were any competition difficulties on this coding ordinance."

NATIONWIDE RESEARCH IN CATTLE REPRODUCTION. A REVIEW

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During the past year or two, several progress reports on cattle reproduction have appeared. The review by Willett *(37)* gives a comprehensive historical account of the development of the subject. One by Roubicek *et al. (29)* is primarily intended to apply to range cattle, but much work with dairy cattle is included, and it gives a more detailed account of contemporary work than does the former paper. A general report of the work of the past 10 yr. in the northeastern states has been published *(7)*. Since these accounts are available, this review will not attempt to survey published work but will be confined to a discussion of trends and to make such suggestions as occur to the writer. Several years ago, Sykes *(31)* gave a review of the problem as it existed then, and his suggestions have formed the basis for much subsequent and valuable work, particularly in the northeastern region.

Arrangements for the exchange of information by workers in the field are well-organized. The northeastern and the western regions have Technical Advisory Committees that meet annually. These meetings enable the members and guests to review the work in progress and to make suggestions for its continuance. These committees function as part of the machinery of the Research and Marketing Act (1946) and have limited budgets for the encouragement of cooperative work on a regional basis. The northeastern project (NE 1) included work upon infective diseases as well as other aspects of the subject. It is now in its last year, but it is to be continued as two separate projects, and committees are now arranging programs, one (NE 40) dealing with pathological aspects and one (NE 41) with nonpathological ones. The western project (W2) was recently reorganized as W49. This new project emphasizes work upon the

basic nature of functional infertility. In addition, a nationwide meeting that brings together workers interested in all classes of livestock is held in alternate years. The proceedings of the first two meetings, at Iowa *(10)* and Michigan *(28)*, have been published. The 1957 meeting was held at Colorado A & M College and the proceedings of this conference also are to be published.

Almost every station has in progress one or more projects concerned in some way with bovine infertility but, in many of them, the relationship is confined to the accumulation of data incidental to the main object of the work, or with the diagnosis and treatment of such cases as arise in the station herd. They do not represent systematic work in the subject. Nevertheless, there is an increasing measure of interest in the field and, as more trained personnel become available, more serious attempts are being made to work out the basic physiology of the organs of reproduction and to improve fertility by applying the new findings.

Lifetime Efficiency

Little information on breeding efficiency can be gained from feeding and management experiments of short duration, because it takes time for most changes to make themselves felt in the reproductive tract. Where differences in feeding are involved, the reproductive organs, if they work at all, tend to work at a high degree of efficiency, especially as the developing embryo has a high priority for the available nutriment *(35)*. It draws upon the maternal system, if supply is short. Long-term experiments are necessary to uncover the effects of chronic deficiencies. Little is known of the effects of various forms of management and of feeding upon lifetime efficiency. The indications are that both may be varied within wide limits without affecting this *(7)*.

Already, there is a trend for more emphasis to be placed upon this question of lifetime reproductive efficiency. In Europe one is impressed with the high proportion of elderly cows in the barns, seemingly far more than there are in the United States. Is this due to a greater effective longevity in Europe, or is it apparent only because of a lower rate of culling for low production? Do we expect too much of our cows and wear them out too quickly? Does a high rate of production affect their reproductive efficiency? There is an increasing body of evidence that reproductive efficiency does not vary as the production level changes *(5, 8)*. But this does not necessarily mean that the cow reproduces equally efficiently at all levels of her own productive capacity. The USDA Cooperative Input-Output experiment *(19)* revealed no difference that could be attributed to feed level. This experiment lasted 2 yr. A similar experiment conducted over the lifetime of the cows would be needed to answer the question fully. Accurate records of feed and labor costs should be kept, to decide whether any prolongation of life, if it appeared, was worthwhile.

A cognate subject is the question whether the rate of growth has an effect upon longevity and upon lifetime production and reproductive efficiency. McCay *et al. (22)* showed that, in rats, slow growth is related to longer life. In this experiment the slow rate was carried to extremes; it was too slow to be applied to cattle as a practical measure. In addition, there was no measure of the value of the days of life gained. These problems are now being investigated at Cornell and at Pennsylvania, where dairy heifers and bulls have been raised at various rates of growth. The effects upon lifetime efficiency are being studied.

Nutrition and Management

In the field of nutrition, an outstanding gap in our knowledge is whether excessive fatness is a cause of sterility, either in the bull or in the cow. The question has never been put to an experimental test. Investigations abroad have suggested that reproductive failure is often due to a disturbed balance between calcium and phosphorus in the diet, especially to a wide calcium-to-phosphorus ratio *(17)*. The evidence presented in support of this view is not conclusive and the results obtained may depend on other conditions. It will be difficult and expensive to conduct a critical experiment that will settle this question, and suitable situations in which the problem may be studied in the field may be hard to find. Some pilot work with laboratory animals might help solve it.

It is most unlikely that shortages of any individual elements or compounds in the feed will cause reproductive failures, unless the supply is so short that the animal shows symptoms of the deficiency. The amounts of dry matter in the ovum and in the semen are so small that they do not represent a drain upon the available nutrients. Also, the rate of embryonic and fetal growth in cattle is so slow that the requirement for this purpose is relatively small and, as has been mentioned, the priority is high. However, it is possible that reproduction might be adversely affected by the presence of unusual substances in the feed. The existence of estrogens in certain forages is known to impair reproduction in sheep by upsetting the normal hormonic balance *(6)*. Possibly, some plants may contain substances that block the action of hormones or other substances necessary to normal function. Several instances of reduced reproduction in species other than cattle may be due to causes of this nature *(12)*.

Much more work needs to be done on the effects of management factors upon economic longevity and lifetime reproductive efficiency. Such work is necessarily time-consuming, but

it need not be expensive if it is carefully arranged. In animal husbandry research of this type, the economic implications are more important than the biological ones, so that the experiments need not be carried to extremes. Extreme experiments should first be done with laboratory animals, because biological data can usually be gathered more efficiently and faster in experiments involving small animals. Pilot tests of this kind often will help settle the limits that should be imposed in an experiment with larger animals, designed with the view of studying possible economic advantages.

Genetics

Population genetic studies have shown that, in this country, inheritance plays but a small part in the total infertility problem *(11, 26)*. The restriction must be made that this statement applies only to those heifers and cows that have shown themselves capable of conception. The rate of conception is modified by heredity in but few animals in the entire cow-population. There is still much work to be done on the impact of heredity upon absolute sterility. Any cases in which the occurrence of inherited factors is suspected should be closely analyzed, and both the nature of inheritance and of the defect should be noted. Careful analysis, too, should be made of any blood lines that are suspected of low fertility. Particular attention should be paid to the probable causes of infertility in individuals in these lines. This is a field of investigation that has scarcely been touched.

Pfau *(24)*, and other geneticists before him, has raised the question whether it is wise by treatment to preserve animals with low reproducing ability for further reproduction. He points out that a genetic gain produced by eliminating detrimental genes is permanent; whereas, preservation of such genes by enabling their possessors to reproduce may lead to the need for more treatments in subsequent generations. The argument for treatment has based itself upon the fact that only in a few instances has heredity been demonstrated, whereas an overwhelming number of conditions of relative infertility are not inherited. In the present state of our knowledge, the writer judges that the risk involved in preserving deleterious genes by treatment is small. The demonstrated inheritance rate is so low and, ironically enough, the prospects of successful treatment so slight, that the risk is minute. If a condition were found, clearly inherited but susceptible to treatment, a conflict might arise between immediate individual interest and the benefit of the breed as a whole. Most breeders would take the longer view.

Disease

At present, methods of curing animals infected with the more serious diseases of reproduction are almost entirely lacking. Brucellosis, for instance, can not be treated by any known method but, fortunately, it can be prevented almost entirely by the use of calfhood vaccination. The search continues for strains other than Strain 19 for vaccination. The metabolism of the causative organism and the immunogenic properties of its antibodies are under study at three experiment stations. More effective methods of diagnosing the disease are being sought widely and one station reports that an attempt is being made to differentiate between natural and vaccination blood titers.

Diagnosis and treatment of trichomoniasis is under study at a few experiment stations and in the Federal Agricultural Research Service. The life history of the organism is also engaging attention.

Vibriosis is attracting a good deal of attention. Several stations and the Agricultural Research Service are studying the morphology and metabolism of the *Vibrio* organism. They are developing improved methods of identification and of differentiating between pathogenic and nonpathogenic strains. At one station, diagnosis, treatment, and transmission in the bull is receiving most attention; another reports an experiment in which a usable and effective vaccine or other preventive agent is being sought and tested.

Infection with *Vibrio* is refractory to treatment in the cow, but the use of insemination, employing semen treated with antibiotics, has proved an effective method of preventing its spread. The fact that cows recover spontaneously after they have been infected is a promising feature for further investigation. What is the nature of the recovery process? Is is sufficiently permanent to confer immunity against reinfection? Could antibodies, if they have been formed, be transferred to calves?

Leptospirosis as a cause of infertility is also under investigation. Special attention is being paid to the possibility that it may be spread through the bull or by artificial insemination.

When abortions occur, it is frequently possible to rule out all the known causes. Then why this large residue of undetected causes? So far, no one has been able to find a major cause besides those already mentioned. A few are due to leptospirosis and there is reason to suspect a virus as a further cause. Attempts to find such a virus have been tantalizing at times. Something recovered from abortuses occasionally survives in embryonated eggs for a few passages, then disappears. Whether these partial results represent the presence of a pathgenic agent is open to question. Possibly, a shift of search methods to tissue culture techniques, using endometrium or mammalian embryonic material as the medium, might yield greater success. One also wonders whether, in some instances, bacteria that are known to be individually relatively harmless may exert a

synergistic action and thus cause damage sufficient to produce abortion.

In all forms of infertility, whether or not due to known organisms, we need more information on the development of symptoms, especially on the nature of changes in the histology of the organs concerned. Perhaps endometrial biopsy may be developed to a greater usefulness when such knowledge becomes available.

As a by-product of experimental work on the effects of management and nutritional differences we may, perhaps, expect to find out whether certain practices contribute to, or limit, infection with specific organisms.

Infertility in the bull is almost a closed book. Histological examination of the testes and accessory organs of bulls of low fertility should be made. This should be a routine matter for bulls discarded from artificial-breeding studs because of low fertility. Perhaps the most vulnerable or sensitive part of the spermatogenic process or of semen production thus could be found. At least, this would provide a basis for subsequent work. One difficulty in establishing the cause of male infertility is that the epididymis is a reservoir for spermatozoa. It may take a month or more to exhaust the reservoir's supply, so that a cause of sterility may be active for some time before it is detected. A febrile condition possibly may cause temporary infertility some time after it occurs.

Artificial Insemination

Many experiment stations report work in progress on artificial insemination. Much of this work consists of a testing by each station of the newest combination of antibiotics or of diluter to be announced elsewhere. The research literature is saturated with reports of work of this nature. Not much basic work is in progress. A few stations are responsible for most of the advances that are made. Several of these are working on the possibility of preserving spermatozoa by drying in some way, and on methods of preservation and storage at room temperature. Results in the latter field, using methods of immobilizing the spermatozoa, seem to be promising *(30, 32)*. Very litle work is in progress on the metabolism of spermatozoa. Yet a knowledge of this aspect might lead to vastly improved methods of handling them.

It is difficult to visualize a function for the fluids from the accessory glands in promoting fertility, because in a normal ejaculation the spermatozoa enter them at the last moment. When the semen is deposited in the female tract, presumably the spermatozoa escape from it at once. The successful use of diluters that resemble the accessory secretions in physical properties but not in chemical composition, likewise suggests that their importance may be exaggerated. However, our knowledge of the composition of these fluids is rather limited. One does wonder whether seminal fluid from a highly fertile bull would improve the fertilizing power of semen from a bull of low fertility.

The whole question of rate of sperm travel *(33)* and of maturing factors for spermatozoa *(9, 38)* should be reopened, and current views, resting as some of them do upon single reports, checked. Would it be possible to feed a bull with radioactive material, some of which would be built into the spermatozoa? Could their radioactivity then be used to track them through the genital organs, either by the use of Geiger counters or by radio-autograph? The rate of sperm formation possibly could be estimated in the same way.

Embryonic Mortality

Embryonic mortality occasions considerable loss, and several stations are investigating the causes. It seems to be clear that good bulls fertilize a high percentage of eggs, and that losses from embryonic deaths cause most of the wastage. In bulls of low fertility, failure to fertilize as many eggs is an additional cause of failure *(4)*. When bulls of low fertility are used, treatment of the semen with antibiotics increases their nonreturn rate *(2)*, an effect observed to a much smaller extent in highly fertile bulls *(1)*. Does the addition of antibiotics increase the fertilization rate, or reduce the embryonic mortality, or both? Evidence is availible that shows a decrease in embryonic mortality *(13)*. Would the addition of specific disease factors to the semen of highly fertile bulls reduce its fertilizing power or increase the subsequent wastage? The problem is whether disease affects fertilization or the life of the embryo. If the latter, is it due to an effect upon the uterus or directly on the embryo? Probably, if semen of low fertility but without disease organisms in the fluid were used, the results of these experiments would differ.

When an embryo dies from a cause other than disease, the question arises whether the cause is in the maternal organism or in the embryo. If it is in the embryo, what causes death? Is it a failure of development in one or another of the tissue or organ systems? In order to answer these questions, detailed reference charts depicting the normal processes of development and implantation are necessary. At present these are lacking, but the gap is being filled as material becomes available *(15)*. This type of work is expensive because it is essential to obtain the material from normal cows. If it is obtained from cows culled for infertility, it may not represent the normal course of development. The expense of providing a large series of reference pictures or photographs is beyond the resources of any journal and of most experiment stations. Special provision should be made for publishing important, but expensive, bulletins of this nature. Why should not a portion of the money appropriated under the Research and Marketing Act be set aside for this purpose? Another use-

ful publication would be one giving in considerable detail the normal cyclical changes in the reproductive tract. These should be referred to ovulation time, not to the time of heat, so that a new series would be necessary to provide the material. Duration of heat and its accurate measuring are too variable for a series based on time of heat to be entirely trustworthy, as the writer has found.

More needs to be done on the problems of timing services. Most of our timing charts are related to the heat period, not to ovulation. There are some reports of surprisingly high conception rates at the extremes *(3, 34)*, and the whole question should be reopened. In this work, the exact time of ovulation should be ascertained, and the fertility percentage curve related to this as well as to the heat period. Some cases of difficulty of conception in cows may be related to ovulation at other than the modal time. Oone wonders whether spermatozoa from highly fertile bulls fertilize more eggs than those from bulls of low fertility, because they survive for a longer time in the female tract. The effect of the act of mating upon ovulation time needs reinvestigation *(21)*.

Control of Ovulation

An important step will have been taken when it becomes possible to bring the ovulation mechanism under control, so that this event can be made to take place at the will of the operator. Present indications are that, although ovulation is spontaneous in the cow, it is brought about by a secretion from the hypothalamus which travels to the anterior pituitary by way of the hypophyseal portal blood system *(16)*. The pituitary then secretes luteinizing hormone which induces rupture of the ripe ovarian follicle. The possibility of control seems to reside in the action of drugs upon the hypothalamus. But many details of the process remain to be worked out. Although the cow has the advantage over laboratory animals, that the results of treatment may be determined with a fair degree of accuracy by rectal palpation of the ovary, she is too expensive for large-scale work with little-known factors. The general control first should be worked out in smaller animals. One line of work should be directed towards finding the essential difference between the spontaneous and the induced ovulators. Another that might profitably be explored is to take advantage of the fact that some clutch-laying species of birds continue to lay if they are prevented from building up the normal clutch number. There seems to be a chain—exteroceptor, hypothalamus, anterior pituitary, ovary—which should be susceptible to detailed analysis. What causes the abrupt transition from laying (LH) to broodiness (prolactin) when the clutch is completed? When the factors involved in the chain are more certainly known, it should be easier to test them in the cow and to work out a method of controlling the process.

The theta wave of the electroencephalogram affords us an opportunity to gauge the activity of the region of the brain in and around the hypothalamus, the region of greatest immediate interest. Some study of these waves when the hypothalamus is stimulated by external factors has been made *(25)* and more use should be made of this approach. Also, when the animal is exposed to light flicker of the same frequency as the theta wave, symptoms, sometimes epileptoid, are produced that appear to be somewhat individual. Possibly, some application of this type of experiment might give us a method of sorting out bulls that would not stand up to the conditions imposed in artificial-breeding studs.

Hormone Assay

So far, all attempts to work out assay methods for sex-steroid hormones applicable to the urine or blood have been unsuccessful. This is largely due to the small amounts of these hormones that are circulating, a condition related to the great sensitivity of the bovine to these substances. It may also indicate a very rapid turnover of active steroids. Nothing is known, as yet, of their metabolism in cattle. Possibly, further applications of paper chromatography methods in this type of investigation might meet with some success. Radioactive tracer techniques are indicated to work out the rate of turnover and the metabolic pathways. The bile has been rather neglected in this respect, and it is probably a major pathway of excretion (see *27*).

Steroid chemistry is tricky; only a small change in configuration frequently makes a large difference in biological activity. The aid of one or more organic chemists with experience in both steroid chemistry and in the use of radioactive materials should be enlisted to help solve the problems outlined. We can not expect to make much progress in treating the "functional" sterilities until we are able to assay the kinds and quantities of hormones in circulation and use, so that correct balances may be restored. More accurate assays of the anterior pituitary for each of the gonadotrophic hormones, separately, in known physiological and abnormal states, are needed to indicate the secretion patterns of these hormones. Some of these suggestions await general advances in this field before applications can be made to cattle. The relationship between gonadotrophic hormone secretion and the histological picture of the anterior pituitary is also worth further study. Some progress in this direction already has been reported by Jubb and McEntee *(20)*.

Mass therapy of infertile cows with hormones has failed to yield results *(14)* but, as our knowledge of endocrine action increases, it should be possible to intervene with success in selected cases. To do so will require a deeper insight into the normal processes than we now have.

Functional Sterility

The so-called functional sterilities are still problems. Nymphomania is not understood and, at present, little or no work is being done on it although some is planned under the NE40 and 41 projects. The uterine physiology does not suggest the action of estrogens. More work upon the development of the condition, particularly as regards the ovaries and the uterus, is needed. How does the progress of the condition, when it is artificially induced by estrogen implants, resemble its natural development? What conditions predispose an animal to an attack? Is the observation *(36)* of an increase in gamma globulin in induced cases a matter of general significance? Will the incidence of the condition increase if a herd is bred along lines that assume that inheritance is involved? Can the condition be made to develop artificially in the absence of the uterus or is it a result of an endometrial overstimulation?

Persistent corpus luteum is another functional sterility that is inadequately understood. A diagnosis of this condition is made all too frequently in cases of anestrum, because ovulation without heat is more frequent than has been realized, as Hurst *(18)* has shown. The condition should not be diagnosed unless the same corpus luteum can be shown to persist as a functional body through at least two cycle-lengths. When the condition does exist, one would like to know the uterine picture as it might offer an explanation of the cause.

The occurrence of heats during pregnancy is a phenomenon that has not been explained. What is the ovarian picture when the condition occurs? In the condition of ovulation without heat, just referred to: Is this due to low sensitivity to estrogens or to their low output?

The cause of the freemartin condition is still obscure. Attempts to modify the gonads of embryos in laboratory animals by injecting the pregnant female with steroid hormones have failed *(23)*, but the method has not been tried in the bovine. Laboratory animals do not ordinarily display the freemartin condition, so that the negative results obtained when they were subjected to the treatment do not necessarily mean that the present hormone theory of its cause is faulty. There are also no reports upon the physiological and histological condition of the anterior pituitary in the freemartin.

Summary

This paper reviews the present trends in research on dairy cattle reproduction in the United States. Gaps in our knowledge are pointed out and suggestions made for work to fill them. A major need is for further work on the study of lifetime reproductive efficiency and the effects of management practices on it. The influence of heredity on the incidence of absolute sterility needs more study. In the field of infectious diseases, search should be pressed for unknown factors, especially for viruses and other subbacterial organisms. Little is known about male infertility. In artificial breeding, the role of the semen plasma needs elucidation. It is an open question whether disease affects fertilization of eggs or early embryonic mortality. Some progress is being made, at last, in mapping the early development of the calf. Timing of service for the greatest fertility should be related to ovulation time more closely than is possible at present. The problem of ovulation control, involving the neuroendocrine mechanism, should receive much more attention. Radioactive tracers should be used in the study of sex-steroid hormone metabolism. "Functional sterility" is still an obscure field that needs further investigation.

Conclusion

In the planning and interpretation of experiments with cattle, we are up against a statistical difficulty in the evaluation of the results. Surely, enough information is available to enable the statistician to tell us how many cows are needed to establish with reasonable significance whether a difference of, say, one-half service per conception between treatments is due to the treatment. Or, how many bulls, randomly chosen, are needed to give reasonable significance to a 1% difference in nonreturn rate. Such information is overdue, and it would be most useful in planning work in which the effects of various modifications of conditions are measured in terms of reproductive performance. Similar information is needed by the pathologist so that the significance of results obtained in experimental testing of therapeutic agents or methods of treatment can be gauged. In these situations, spontaneous recovery rates in animals with definite symptoms are needed for comparative purposes. Finally, what advantages over the spontaneous recovery rates are necessary to make treatment financially worthwhile at different yields?

Much of the progress in our knowledge of human physiology was obtained by careful study of abnormal conditions. These led to the recognition of the functions of the endocrine organs and to the nature of the balances necessary for their orderly function. The writer has still to see the day when an experiment station or college has the fortitude to use the better herds in its neighborhood for teaching purposes and to concentrate on collecting all the abnormal livestock it is possible to find. He is sure that an intensive study of these animals by the station staff would rapidly pay ample dividends in research, if not in public relations.

In general, it is believed that the present knowledge of reproduction and the possibility of its control in the bovine lags because many of the basic problems are still unsolved.

There is need for much more detailed work upon causes and mechanisms. Study of these

requires special skills of a high order. The writer endorses Willett's *(37)* view that men of this type should be brought in to develop those areas where the deficiency of expert knowledge is most apparent.

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OUR ASSOCIATION

GOLDEN ANNIVERSARY OF EVENTFUL MEETING AT CORNELL UNIVERSITY

G. M. TROUT[1]
Department of Dairy, Michigan State University, East Lansing.

Fifty years ago, July 22 and 23, 1908, the two-year-old "Official Dairy Instructors Association" (later to become the American Dairy Science Association) held its annual meeting on the Cornell University campus, Ithaca, New York. Perhaps few meetings of the Association since then have equaled this one in the percentage rise in attendance from previous meetings, and in the number of those destined to become enshrined in the halls of dairy fame.

Membership had increased from the 17 charter members at Urbana, 1906, to 69 members from 26 states, the District of Columbia, and Canada. Forty-seven posed for a picture, which later was to become of historical interest. Eleven of those attending the third annual conference at Cornell in 1908 were to attend the 29th annual meeting in 1934, again at Cornell. At least five were present again at the 45th Cornell meeting, 1950.

Of those present at the 1908 Cornell meeting, Lee, Thom, Doane, Larson, C. W., Hunziker, Ross, Washburn, Webster, Hayden, White, Haecker, Rasmussen, Larson, C., Erf, Eckles, Pearson, Savage, Van Norman, Van Slyke, Miyawaki, Stocking, Publow, Nystrom, Dean, Guthrie, Frandsen, Armsby, Farrington, Woll, Rawl, and others, were to carve niches for themselves in dairy science and have their names revered among agricultural leaders of America during the next 50 years. Interestingly, no less than three members present, Hunziker, Eckles, and Haecker, were to be named among the all-time "Ten Master-Minds of Dairying." At least 16 were to become authors of dairy texts.

Certain characteristics destined this third conference to become of more-than-routine historical significance. In the first place, membership was up, attendance was relatively high, and interest was good. Secondly, the president, Dr. R. A. Pearson, serving his second year, confined his address to a few remarks, emphasizing the need of interesting, competent young men in dairy industry, in which good positions were awaiting incumbents. Thirdly, the program consisted not of set, topic papers by individuals, which featured the first meeting, but of thorough, thought-provoking, action-stimulating committee reports. However, the committee reports that formed the kernel of the second annual meeting at Chicago, October, 1907, must not be deprecated but, judging from the discussion of them, these reports did not carry the substance and finality of the thinking (and conclusions) of the committee members as did the (committee) reports of the Cornell conference. Fourth, these committee reports were to become, in large part, the guide for the thinking of the members of the Association for years to come. In fact, the "Report of Committee on Experimental Work: Production. Some Suggestions Regarding Methods of Conducting Experiments with Dairy Cows" by W. J. Fraser, Chairman (the founder of the Association and who was not present at the Cornell meeting), C. H. Eckles of Missouri, and H. H. Dean of Guelph laid down some basic principles in dairy cattle feeding experiments which were grasped then by the young scientists and used even to this day. With possibly one exception, full, complete reports were presented by the following committees: "Dairy score-card"; "National dairy register of merit"; "Our relations to the National Dairy Show"; "Courses of instruction"; "Experimental work: Production"; "Experimental work: Manufactures"; "Standards for dairy products"; "Extension work"; "Extension work in the United States"; "Official testing"; "Cooperative work," and "Cow test associations. "

Already, the leadership of the 2-year-old "National Association of Dairy Instructors and Investigators" was manifesting itself in the development of score-cards, standards for products, rules for dairy cattle judging contests, testing and identification of dairy cows, instruction and research, and even in dairy extension, which was not to be recognized and provided for officially by the United States Government until the passage of the Smith-Lever Act of 1914.

As a result of the Cornell conference, and through the activity of a segment of the new organization, the first college dairy cattle judging contest was set for December 3, 1908. This contest has been held almost every year since then, and has had a profound influence on the training and stimulation of future dairy leaders.

Unfortunately, the good Ithaca conference, at which so much was accomplished for the potential development of dairying, was the last of such official, summer, on-college campus meetings to be held for 19 years. However, yearly official meetings were held with the National Dairy show. On several occasions, unofficial

[1] Historian, American Dairy Science Association.

CORNELL UNIVERSITY—JULY 22ND AND 23RD, 1908

1. Lee, C. E., Urbana, Ill.; 2. Dox, A. W., Storrs, Conn.; 3. Kendall, J. C., Manhattan, Kansas; 4. Thom, C., Storrs, Conn.; 5. Ayres, H. L., Ithaca, N. Y.; 6. Willoughby, C. L., Experiment, Georgia; 7. Dexter, W. H., Washington, D. C.; 8. Doane, C. F., Washington, D. C.; 9. Campbell, P. A., Orono, Maine; 10. Larson, C. W., State College, Pa.; 11. Hunziker, O. F., Lafayette, Indiana; 12. Little, E. M., Lincoln, Nebraska; 13. Beach, C. L., Storrs, Conn.; 14. Ross, H. E., Ithaca, N. Y.; 15. Washburn, R. M., Burlington, Vt.; 16. Webster, E. H., Washington, D. C.; 17. Hayden, C. C., Urbana Ill.; 18. Ayer, L. F., Hampton, Va.; 19. White, B. D., Washington, D. C.; 20. Kent, F. L., Corvallis, Oregon; 21. Haecker, T. L., St. Anthony Park, Minn.; 22. Rasmussen, F., Durham, N. H.; 23. Whittaker, G. M., Washington, D. C.; 24. Larson, C., Brookings, Dakota; 25. Erf, O., Columbus, Ohio; 26. Merrill, L. H., Orono, Maine; 27. Jarvis, J. D., Lafayette, Ind.; 28. Eckles, C. H., Columbia, Missouri; 29. Billings, G. A., Washington, D. C.; 30. Pearson, R. A., Albany, N. Y.; 31. Savage, E. S., Ithaca, N. Y.; 32. Van Norman, H. E., State College, Pa.; 33. Van Slyke, L. L., Geneva, N. Y.; 34. Miyawaki, A., Manhattan, Kansas; 35. Melick, C. W., College Park, Maryland; 36. Lane, C. B., Washington, D. C.; 37. Stocking, W. A. Jr., Ithaca, N. Y.; 38. Caldwell, R. E., Manhattan, Kansas; 39. Publow, C. A., Ithaca, N. Y.; 40. Nystrom, A. B., Columbus, Ohio; 41. Ferguson, A., Ithaca, N. Y.; 42. Dean, H. H., Guelph, Canada; 43. Guthrie, E. S., Columbus, Ohio; 44. Simpson, O. G., Corvallis, Ore.; 45. Frandsen, J. H., Moscow, Idaho; 46. Wilson, J. K., Geneva, N. Y.; 47. Rawl, B. H., Washington, D. C.

meetings continued to be held biennially on a college campus with the summer meetings of the Graduate School in Agriculture.

Ironically, the National Association of Dairy Instructors and Investigators, founded so well at Urbana in 1906, and relaunched with full steam at Cornell in 1908, was due for at least "Seven Lean Years," if not longer. A "dark era" in the life of the 2-year-old association was ahead. During this period, little about the Association seems to have been recorded. The founding of the Journal in 1917 gave the Association new life. The re-establishment of the summer on-college campus meetings at East Lansing in 1927 was the beginning of an upsurge in attendance at the annual meetings of the Association.

The influence of the annual meetings of the American Dairy Science Association such as the one at Cornell, 1908, can be measured only in the lives it has touched. And these are immeasurable. Radiating out from distinguished men such as were gathered at this eventful meeting, were forces destined to leave their imprint on the minds of those within their range. Only after a life of active service may these influences be viewed in proper perspective. Reading between the lines of Amer B. Nystrom's recollections of the 1908 Cornell meeting, after half a century, one gets a hint of the satisfaction and sustaining force which can be had by attending the meetings of the American Dairy Science Association. He writes:

I was a youngster—only 1 year out of college, when I went to Ithaca for the 1908 sessions and I felt very fortunate to be able to attend. Aside from the business sessions, which I don't remember at all, I took in all the lectures and demonstrations on dairying, bacteriology, and general subjects that did not conflict. I particularly remember Dr. W. W. Stocking's talks on Dairy Bacteriology. Also B. D. White's on Market Milk and C. F.

Doane's on Cheesemaking. Dr. Van Slyke also talked on Cheesemaking.

Prof. Oscar Erf, then at Ohio State, was one of the leaders in the Dairy Instructors Association's sessions at that meeting. The photo mentioned above shows him and other officers in the front row, as I recall. I can't remember who the other officers were, but you may be able to figure it out from the picture.

I don't believe Dr. S. M. Babcock was there, but we had demonstrations and talks on the Babcock test, which, as I recall, were given by either Farrington or Woll or both. Then we had a couple of lectures on nutrition by Dr. Armsby. He talked about the energy (calorie) value of feeds and their effects on dairy cow health and production.

Outside of the dairy field, I remember some excellent general talks by Dean Liberty Hyde Bailey which I enjoyed. And one evening Dean Bailey had us all in for a social evening at his home. There he talked to us again, in an entertaining way which we all appreciated.

Perhaps the stimulus engendered by attending the annual Association meetings, as reflected in the above letter, can not be evaluated in its entirety. In time, the attendance at the annual meeting becomes an urge for the dairy scientist. Many of those attending the third annual meeting on the Cornell campus, 1908, welcomed the opportunity to return again and again to the annual gathering. They learned early the meaning of President W. A. Stocking, Jr.'s words spoken at Columbus, Ohio October 14, 1917, "It is a pleasant thing for us to get together each year, and throw off the restraint when we have a chance, and visit informally."

VOLUME 41 June, 1958 NUMBER 6

JOURNAL OF DAIRY SCIENCE

ABSTRACTS OF LITERATURE

W. O. Nelson, Abstract Editor

BOOK REVIEWS

221. The Lipids, Their Chemistry and Biochemistry. Vol. III. Biochemistry, Biosynthesis, Oxidation, Metabolism, and Nutritional Value. H. J. DEUEL, JR. Interscience Publishers, Inc., New York, 1957, 1,065 pp. $25.

The third volume of *The Lipids* deals primarily with the metabolism of fats and completes the discussion of the chemistry (Vol. I) and the biochemistry (Vol. II) of this monumental treatise on lipids. It was completed by Dr. Deuel shortly before his death and was seen through press by some of his many colleagues and friends.

This volume represents a natural sequel to the second volume, The Biochemistry of the Lipids. The latter volume considered in detail the digestion, absorption, and transportation of lipids. The third volume treats the biosynthesis, oxidation, and intermediate changes which are involved in the metabolism of lipids and, in the last chapter, the nutritional value of fats.

The third volume is divided into 14 chapters and contains a total of 4,255 references. The first chapter is composed of a short review of digestion, transport, and storage, which were considered in detail in the second volume. The succeeding chapters present a thorough discussion of the biosynthesis of triglycerides, phospholipids, and fatty acids and their oxidation and metabolism. The questions of the conversion of fat to carbohydrate, the role of acetic acid in intermediary metabolism of fats, and the metabolism of branched chain fatty, hydroxy, and keto acids, dicarboxylic acids, tricarboxylic acids, fatty acid amides, aldehydes, and hydrocarbons are considered. A considerable amount of space is devoted to the metabolism of cholesterol, carotenoids, and vitamins A, D, E, and K. The physiology and biochemistry of the essential fatty acids also received adequate coverage.

As with the two previous volumes, this book is not intended or recommended as a sole text for students of lipid chemsitry. It is most useful as a reference book and for review for the student, as well as the research worker in lipids.

The publication of this third and final volume completes a unique work in the available literature regarding the lipids. To derive the full benefit of the information in this work it is necessary to have access to all three volumes. The reader of Volume III will find himself frequently referred to Volumes I and II for a completion of the required information. In these three volumes is stored most of our present knowledge of the large number of compounds which are classed under the term "lipid." P. V. Johnston

222. Dairy Plant Management. PAUL H. TRACY, GEORGE H. ARMERDING, AND HAROLD W. HANNAH. McGraw-Hill Book Company, New York. 439 pages. 1958. $8.50.

This is a textbook that will be of great value in dairy plant management courses for the college student. Also, college graduates and others who are interested in a better understanding of management problems and functions will find it most beneficial.

The authors have brought together information from the many areas of plant management and adapted it to the dairy industry. Heretofore, this information has been available in fragmentary amounts and in widely scattered places. The authors, because of their experience in the dairy industry and in management, have been able to assimilate and present in an effective manner a broad discussion of management problems.

The text follows in very logical order from beginning a business to the planning, building, and operating of the plant; selection and supervision of personnel; office management, with reports and records; purchasing of raw materials, equipment, and supplies; sales, advertising and public relations, and the legal aspects of running a business. The chapters on labor unions, industrial relations, and accident prevention will be of interest to those in industry who are being confronted with these problems for the first time.

The selections on savings plans and developing leadership will be of particular use to college students in preparation for entrance into the industry.

The book meets a need that has been sharply focused in recent years by the growing importance of many related aspects of successful plant management to problems in the dairy industry. W. M. Roberts

DAIRY CHEMISTRY

223. Acetylcholine in milk. V. P. Whittaker. Nature, **181**, 4612: 856. 1958.

This substance was tested by its action on the frog rectus abdominus muscle. The conclusion is drawn that the acetylcholine content of milk is very small and that the contractions of the frog muscle are caused, instead, by the potassium, choline, and histamine content of the milk. R. Whitaker

224. Bacterial oxidation of lactose and melibiose. B. P. Eddy. Nature, **181**, 4613: 904. 1958.

An organism isolated from whale meat has the property of oxidizing lactose and melibiose to lactobionic and melibionic acids, respectively, rather than hydrolyzing these sugars to simpler sugars. R. Whitaker

GENETICS AND BREEDING

225. Separations of bull spermatozoa carrying X- and Y-chromosomes by counter-streaming centrifugation. P. E. Lindahl. Nature, **181**, 4611: 784. 1958.

Female-determining spermatozoa are injured more readily than the male-determining type when sedimented by counter-streaming centrifugation. Mechanical stress is given as the probable cause. Operating the centrifuge at moderate speeds is suggested as a means of increasing the ratio of females to males resulting from artificial insemination.

R. Whitaker

ICE CREAM

226. Sixth annual Ice Cream Field survey of industry trends. Anon. Ice Cream Field, **71**, 2: 66. 1958.

According to this survey, super-markets and grocery stores sold 54.93%, restaurants 13.50%, and drug stores—which at one time were the largest outlet—8.10% of the ice cream in 1957. A long list of special flavors is presented in the survey.

The average prices at retail level were factory pt., 28¢; hand-pack pt., 45¢; factory qt., 56¢; hand-pack qt., 84¢; packaged ½ gal., 95¢, and super-market ½ gal., 79¢.

An increase in the use of plastic containers in 1958 is predicted.

Newspaper and point-of-purchase advertising constitute slightly over 50% of the ice cream advertising budget. W. C. Cole

227. Stabilized bodied milk and frozen milk products comprising carboxymethyl dextran. H. A. Toulmin, Jr. U. S. Patent 2,834,684. May 13, 1958. 3 claims. Offic. Gaz. U. S. Pat. Office, **730**, 2: 461. 1958.

An ice cream stabilizer consisting of dextran combined with 2 to 3 carboxymethyl groups per anhydroglucopyranosidic unit of dextran is described. R. Whitaker

228. Unrefrigerated dairy cream is studied. Anon. Ice Cream Field, **71**, 1: 50. 1958.

Sugar-preserved dairy cream that keeps without refrigeration is a new product developed by Dr. Tamsma and Dr. R. W. Bell, of the United States Department of Agriculture, Washington, D. C. It contains about 40% butterfat, 10% milk-solids-not-fat, and 30% sugar.

It can be used as an ice cream ingredient and also may be used in other ways.

W. C. Cole

NUTRITIVE VALUE OF DAIRY PRODUCTS

229. On the feeding of solid foods to infants. Report of the Committee on Nutrition, American Academy of Pediatrics. Pediatrics, **21**, 4: 685. 1958.

Before 1920, babies seldom received any food other than milk until about 1 yr. of age. In the 1920's reports appeared on the early introduction of solid foods, even to the point of replacing milk entirely. Solid foods were added to the diet early, to supply iron, certain vitamins, and possibly other food factors and to acquaint the baby at a susceptible age with a varied dietary pattern.

In 1937, the American Medical Association stated that the feeding of strained fruits and vegetables at an age of from 4 to 6 mo. was generally favored by pediatricians. In a survey conducted by the Quarterly Review of Pediatrics in 1954, 88% of 2,000 pediatricians who answered stated that they began feeding solid foods before the age of 3 mo.; 66% reported starting before 8 wk. Recently, successful feedings with cereal starting on the second or third day, strained vegetables at ten days, meats at 14 days, and strained fruits at 17 days, have been reported.

The Committee, after studying all such reports, including the possible specific benefit from early feeding of meat from its hemoglobin-producing properties, and taking into consideration the possible physiologic and psychologic factors, finds that proof is lacking of any benefits from starting solid foods earlier than about 3 mo., provided the milk formula is supplemented with vitamins C and D. Depending on the individual baby, the introduction of solid foods may be slightly earlier or later. Thus, the prematurely born baby may not be

ready for foods other than milk until well beyond the age of 3 mo.

The Committee states that a baby on a properly constituted cow's-milk formula containing 20 calories per ounce receives 110-120 calories/kg/day. The addition of solid food in the first few weeks of life may well have the effect, through satiety, of lessening the volume intake of milk. Although the total caloric requirement might be adequately met, the feeding of solid foods nutritionally inferior to milk, at the expense of milk, could result in worsening rather than bettering the nutritional state of the infant. Frank E. Rice

230. Food allergy and food intolerance in relation to the development of good eating habits. E. L. PRATT, Dept. Pediatrics, Univ. of Texas School of Medicine, Dallas. Pediatrics, **21**, 4:642. 1958.

This is a review and commentary on our knowledge and lack of knowledge of food ingestants that may be causative factors in allergy and such syndromes which many times are diagnosed incorrectly as allergy. Some of the latter could be better designated by the broader term, "intolerance."

The author emphasized the need for carefully planned research to find objective tests to determine when a dietary constituent is truly an etiologic factor in allergy. Skin tests are confusing and unreliable. Food substitution is not a good criterion. One investigator reports that withholding cow's milk from babies when there were family histories of allergies resulted in fewer "major" allergies. However, withholding milk in the neonatal period did not reduce the likelihood of babies later developing allergy to milk.

The author states that unless allergy, or intolerance to milk, is definitely established, tampering with infants' diets is not justified, and may have immediate and distinct effects on nutrition, and on the development of good life-long eating habits. Frank E. Rice

231. Essential fatty acids and infant nutrition. A. E. HANSEN, Univ. of Texas School of Medicine, Dallas. Pediatrics, **21**, 3:494. 1958.

Linoleic acid plays a significant metabolic role in infant and animal nutrition. Dietary deficiency is indicated by disturbed skin function—dermatitis, eczematous lesions, etc. When the intake is inadequate, a lower level of di- and tetraenoic acids is found in the serum. However, under the same conditions the content of trienoic acid increases.

An example is given of an infant fed for a period of time on a skimmilk mixture, then transferred to an evaporated milk formula. Blood serum analyses showed that, following the change, the content of dienoic and tetraenoic acids doubled, yet the level of trienoic acid declined by about one-fourth. In the second feeding period, a definite decrease in the calories consumed per unit of body weight was observed, indicating a better utilization of the food. Rates of weight gain were about the same in the two feeding periods. Frank E. Rice

232. Infant feeding—The evaporated milk chapter. FRANK E. RICE, Minnesota Medicine, **41**, 4:266. 1958.

General acceptance of evaporated milk as the basis for baby-feeding formulas by the medical profession started with 1929, the year two prominent pediatricians independently published observations on its use with a large number of babies. During the 1930's clinical and biological research revealed no important loss of nutrients as a result of evaporation. Other factors which appealed to the medical profession, and which contributed to the acceptance of evaporated milk, included lowered curd tension, homogenized condition of the fat, fortification with vitamin D, uniformity, universal availability, and relatively low cost.

A recent survey shows that at present two-thirds of the babies born in U. S. hospitals are discharged on a bottle formula of some kind. In these formulas evaporated milk is prescribed more than any other form of milk, including the special proprietary preparations.

Frank E. Rice

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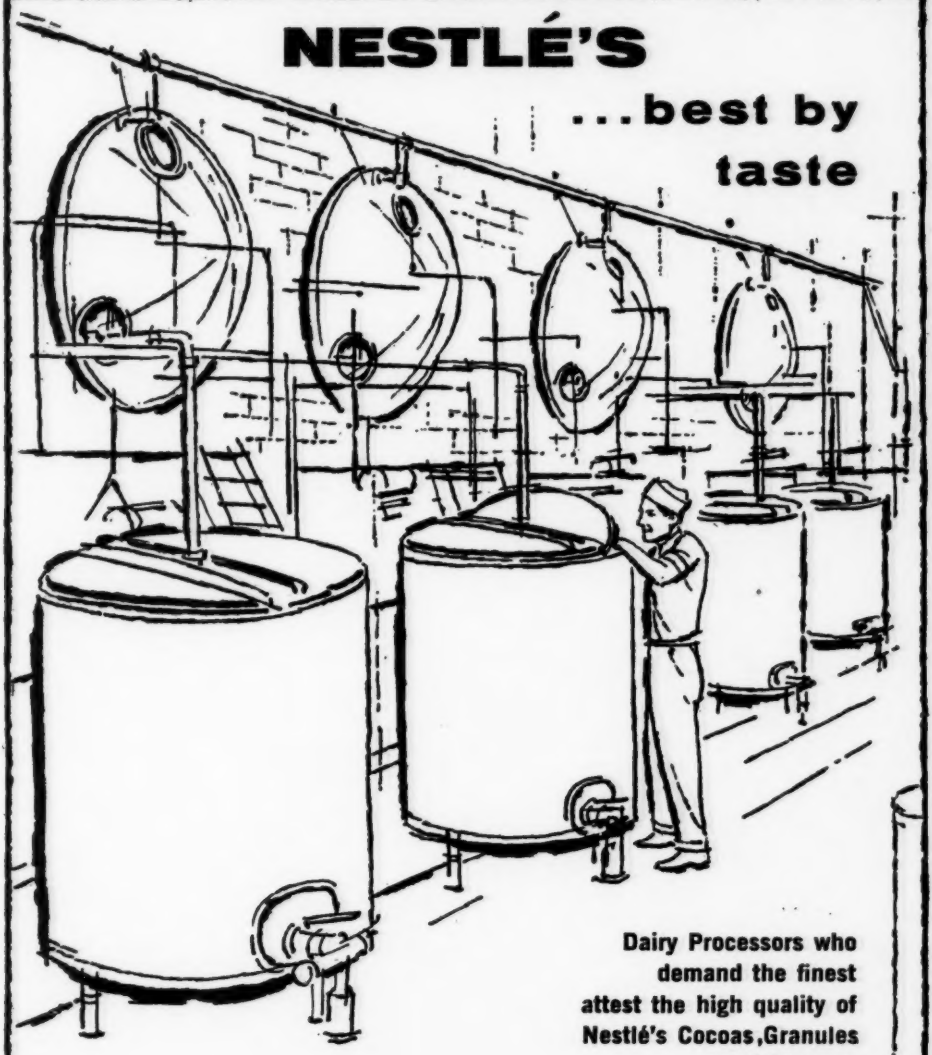

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